Gonadal steroids, brain, and behavior: role of context

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The nature and extent of the impact of gender and reproductive function on mood has been the subject of speculation and controversy for centuries. Over the past 50 years, however, it has become increasingly clear that not only is the brain a major target of reproductive steroid hormones, but additionally, the steroid hormones, as neuroregulators, create a context that influences a broad range of brain activities; ie, neural actions and resultant behaviors are markedly different in the presence and absence of gonadal steroids. In turn, the actions of gonadal steroids are themselves context-dependent. Thus, even where it can be demonstrated that gonadal steroids trigger mood disorders, the triggers are normal levels of gonadal steroids (to be contrasted with the mood disturbances accompanying endocrinopathies), and the mood disorders appear only in a subset of susceptible individuals. The context specificity and differential susceptibility to affective dysregulation seen in women with reproductive endocrine–related mood disorders are undoubtedly important underlying characteristics of a wide range of psychiatric disorders in which the triggers have not yet been identified. Consequently, reproductive endocrine–related mood disorders offer unparalleled promise for the identification of those contextual variables that permit biological stimuli to differentially translate into depression in individuals at risk.

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This issue of *Dialogues in Clinical Neuroscience* focuses on depression and senescence in women for several reasons. First, mood disorders linked to reproductive endocrine change in women (eg, premenstrual syndrome [PMS], postpartum depression [PPD], and perimenopausal depression [PMD]) are clinically significant: they are prevalent and attended to by considerable morbidity. Second, it is now clear that reproductive steroids are important regulators of virtually every aspect of brain organization and function, from neural differentiation and migration to intracellular and intercellular signaling to neuronal (and glial) survival and death. Simply put, these steroids create a context such that the brain functions differently in their presence and absence. Third, political objections notwithstanding, it is now clear that reproductive steroids play a critical role in triggering reproductive endocrine–related mood disorders, thus compromising arguments that these disorders do not exist or that the ostensible link to reproductive endocrine change is illusory. Fourth, it is equally clear that reproductive steroids do not, by themselves, cause reproductive endocrine–related mood disorders; ie, women with these disorders are differentially sensitive to levels or changes in reproductive steroids that are without effect on mood in women lacking these disorders.

This differential sensitivity, the fact that people respond differently to the same reproductive endocrine stimulus, is important for three reasons. First, the failure to

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Selected abbreviations and acronyms

AP1	*activator protein 1*
AR	*androgen receptor*
DHT	*dihydrotestosterone*
ER	*estrogen receptor*
fMRI	*functional magnetic resonance imaging*
GABA	*γ-aminobutyric acid*
hCG	*human chorionic gonadotropin*
3α-HSOR	*3α-hydroxy steroid oxidoreductase*
MAPK	*mitogen-activated protein kinase*
PET	*positron emission tomography*
PMD	*perimenopausal depression*
PMS	*premenstrual syndrome*
PPD	*postpartum depression*
T3	*triiodothyronine*

consider this phenomenon has been the keystone of the argument that reproductive endocrine–related mood disorders do not exist. The syllogism is as follows: The effects of reproductive hormones should be similar across individuals; not all individuals have the same behavioral concomitants of changes in reproductive endocrine function; therefore, reproductive hormones have nothing to do with behavior. In other words, if changes in reproductive steroids do not precipitate mood disorders in everyone, they must do so in no one. As will be shown below, this argument is fallacious and serves to obscure rather than clarify. Second, the principle that the response to a biologic stimulus depends upon the context in which the stimulus is administered is generalizable and underlies much of our physiology. Third, by understanding the means by which reproductive steroids can trigger affective change in some but not other women, we will be in a far more powerful position to understand the substrate underlying susceptibility to affective disorder in general, which is better conceptualized as a differential response to a stimulus rather than as a "deficiency" state. The remainder of this paper, then, will address the central question in reproductive psychiatry/neuroscience: how is it that reproductive steroids can trigger a depression, and why does this occur only in some individuals?

Reproductive steroids: modulators of brain function

The observed links between reproductive function and behavior date back at least several millennia to Aristotle, who noted that castration of immature male birds prevented the development of characteristic male singing and sexual behavior.[1] By the end of the 19th century, Brown-Séquard and other "organotherapists" claimed that the administration of ground-up extracts from animal gonads could successfully treat a variety of human mood disorders, including depression and the anergy of senescence.[2-5] In the 1920s and 1930s, the potential mediators of these effects—the steroid hormones estradiol, progesterone, and testosterone (and not sperm, as Brown-Séquard believed)—were isolated and characterized. Forty years later, Jensen and Jacobsen[6] demonstrated that the actions of estradiol occurred through its binding to an intracellular protein, the estrogen receptor (ER), which was isolated and identified 4 years later.[7] The means by which steroids influenced cell function were subsequently elaborated: after diffusing into the cell, the steroid bound and activated its receptor, a transcription factor that could then bind DNA and regulate the transcription of mRNA, which would then be translated into proteins in the cell cytoplasm. In this fashion, reproductive steroids were found to regulate the expression of a variety of proteins of relevance for neural function (eg, neurotransmitter synthetic and metabolic enzymes, neuropeptides, receptors, etc). More recently, advances in neuroscience (reviewed in the accompanying article by McEwen[8]) have demonstrated vastly more complex, broad-ranging, and powerful mechanisms for neural control by reproductive steroids, and have further uncovered several regulatory principles that help explain how a given steroid signal may elicit diverse behavioral responses. One inescapable, overarching principle is that the molecular and behavioral effects of steroids are highly context-dependent.

Cellular context

Data overwhelmingly suggest that the cell is a context that determines the response to a stimulus. First, steroid-activated receptors influence transcription not as solitary agents, but by forming combinations with other intracellular proteins.[9] Some of these proteins, the coregulators, determine whether gene transcription is enhanced or suppressed by the activated receptor. Other proteins, the cointegrators, permit activated receptors to regulate genomic expression through sites (eg, activator protein, AP1) other than the classical DNA hormone response elements, thus expanding the range of genes

influenced by steroids.[10] Many of these proteins are tissue specific, thus helping to explain how ER modulators (eg, tamoxifen and raloxifene) can act like agonists in some tissues (eg, bone) and like antagonists in other tissues (eg, breast).[11,12] Second, different subtypes of the steroid receptors are either coded for on different genes (eg, estrogen receptors alpha and beta)[13] or modified after transcription (eg, splice variants or progesterone receptor isoforms A and B).[14] These subtypes have different distribution patterns in the brain, different affinities for ligands, and very different actions (including inhibition of the actions of other subtypes).[15,16] Third, the relatively slow, "genomic" effects of reproductive steroids have been expanded in two dimensions: time, with a variety of rapid (seconds to minutes) "nongenomic" effects observed; and targets, which now include ion channels and second messengers. Once again, the effect observed depends upon the type of cell examined: estradiol activates the second messenger, mitogen-activated protein kinase (MAPK), in neurons, but decreases MAPK activation in cortical glia.[17]

Metabolic context

As steroid hormones are highly homologous and serve as precursors for one another, the manner in which steroids are metabolized can markedly change the amplitude or nature of the steroid signal. Steroid metabolic enzymes, then, can contribute to the variance in a steroid signal in several ways. First, enzymes regulate the activation and potency of steroid hormones, as seen, for example, with the enzyme (5α-reductase) that converts testosterone into dihydrotestosterone (DHT), an androgen with fourfold greater affinity for the androgen receptor (AR) and fivefold greater stability.[18] Second, enzymes determine the receptor system that is activated, as seen, for example, in the conversion by aromatase of testosterone (acting at the AR) to estradiol (acting at the ER). Third, the metabolism of steroids can facilitate or inhibit the accumulation of metabolites that may be neurotoxic, as seen, for example, with the ability of 5α-reductase to shunt testosterone away from the pathway leading to accumulation of estradiol, which can function as a neurotoxin.[19,20] Fourth, enzymes may produce steroid metabolites that have a completely different neuromodulatory profile from that of the parent hormones, as seen, for example, with the conversion of progesterone to the neurosteroid allopregnanolone (by 5α-reductase and 3α-hydroxy steroid oxidoreductase [3α-HSOR]), a potent modulator of the γ-aminobutyric acid (GABA) receptor chloride ionophore.[21] Finally, since many of the enzymes have multiple steroid substrates, the enzyme activity regulates the relative amounts of different behaviorally active metabolites; for example, 3α-HSOR both inactivates the androgen DHT and produces the neurosteroid allopregnanolone.[22] Not only will different metabolic profiles activate or inhibit different receptor systems, but the consequence of the activation of a given steroid receptor will differ depending upon which hormones are present. Estradiol and cortisol, for example, exert opposing effects on AP1-modulated genes through interactions with the cointegrator CBP/P300.[10] A steroid hormone, then, may produce markedly different effects depending upon its metabolism and the hormonal context in which it is acting.

Developmental/temporal context

Perinatal reproductive steroids create a context that influences (organizes) brain development and the adult behavioral repertoire. Phoenix et al[23] and Gorski et al[24] showed that prenatal exposure of female guinea pigs or perinatal exposure of rats to androgens resulted in enhanced behavioral sensitivity (eg, increased sexual and aggressive behaviors) to androgens administered during adulthood. Thus, differences in early exposure to reproductive steroids created the capacity in adults for different behavioral responses to the same stimulus. The effects of reproductive steroids are also developmental stage–specific. Estradiol, for example, stimulates its own receptor early in development, inhibits it during adulthood, and stimulates it again in the context of brain injury.[25] Modulatory effects of reproductive steroids also differ in old and young subjects (both animals and humans). For example, spine density in the dentate gyrus is modulated by estradiol in old but not young female rats.[26]

These age-dependent effects are particularly of interest given a burgeoning literature describing the ability of reproductive steroids to regulate cell death and survival through effects on cell survival proteins (eg, Bcl-2, Bax), signal transduction (eg, MAPK, Akt), amyloid precursor protein metabolism, and free radical species generation.[17,27-30] Effects on survival operate at both ends of the developmental spectrum. Early effects influence prun-

ing[31] and the shaping of brain circuitry. Modulation of neural and glial survival during aging provides yet another means by which reproductive steroids may influence the susceptibility to neuropsychiatric illness, given the putative role of neurodegeneration in depression[32-34] and its demonstrated role in Alzheimer's disease.

Environmental context

The brain is a nonlinear transform system, in which the response to a stimulus can be altered as a function of past history or present environment. Multiple demonstrations of this process can be found in the animal literature. For example, behavioral sensitization refers to an amplified behavioral response (eg, aggression) to repeated exposure to a pharmacologic stimulus.[35] Two elements of this process are of further interest. First, Antelman has suggested that even without repeated administration, exposure to certain drugs may yield an amplified response upon readministration, simply by virtue of the passage of time.[36] There is a "memory" following exposure that alters the response when the stimulus is re-presented. Second, Post and coworkers have demonstrated that expression of behavioral sensitization may be context dependent, in that the exaggerated response elicited to cocaine in the test cage will not be manifest if, after sensitization is achieved, the cocaine is administered in the home cage.[37] Both past experience and environment, then, may alter subsequent response. One of the most impressive demonstrations of experience (and development)–related alterations in context is provided by the work of Meaney and coworkers. These authors[38] expanded the work of Levine[39] and showed that the separation and handling of rat pups elicited licking and grooming behavior from mothers that differentially and permanently determined the nature of the offspring's response to stressors. Meaney and coworkers then went on to demonstrate in cross-fostering studies that it was the maternal licking and grooming behavior, not the genetic factors, that influenced the licking and grooming behavior (as well as the stress responsivity) of the female offspring, and that the "adopted" licking and grooming behavior and stress responsivity were passed down to subsequent generations.[40] This series of studies, then, demonstrates that maternal behavior can alter the developmental context, such that permanent and dramatic differences in response—from the transcriptional to the behavioral level—are programmed into the offspring. Several studies also demonstrate the exquisite sensitivity of reproductive physiology and behavior to environmental alterations during development. Ward and Weisz[41,42] demonstrated that male offspring of a rat dam stressed during gestation were demasculinized, with lower testosterone levels (on critical gestational days) and deficient adult male mating behavior.[43] Moore et al[44] observed that the size of the sexually dimorphic spinal nucleus of the bulbocavernosus as well as adult male mating behavior were in part determined by maternal licking of the anogenital region of the pup, which in turn appeared to be elicited by androgens in the rat pup urine. Finally, reproductive hormones interact with environmental factors during development to determine the adult behavioral repertoire. Adult aggressive behavior in mice can be attenuated by prepubertal castration; the attenuation, however, is blunted to the extent to which the mouse has already been exposed to aggressive encounters.[45] These examples demonstrate that current and past environments and experience can create a context in which the same hormonal or environmental stimulus may elicit any of a range of behavioral responses.

Gender context

Early hypotheses that the brain displays sex-related differences in structure and function were confirmed by the demonstrations by Pfaff[46] of sexual dimorphisms in rat brain morphology and by Raisman and Field[47] of sex-related differences in the synaptic density of the preoptic area in the rat. There is now an impressive literature detailing sexual dimorphisms at all levels of the neuroaxis, including differences in the following: nuclear volume; neuron number, size, density, morphology, and gene expression; signal transduction; neuronal neuritic branching patterns; synapse formation; and physiological and behavioral response.[48,49] Given the ability of reproductive steroids to regulate virtually all stages of brain development, from neurogenesis to neural migration, differentiation, synaptogenesis, survival, and death,[49] the wide range of brain dimorphisms is not surprising. Nonetheless, the source and significance of many of the dimorphisms are far from clear. For example, while exposure to reproductive steroids is believed to organize (perinatally) or activate (adulthood) most dimorphisms, Reisert and Pilgrim[50] showed that dimorphisms in the course of development of embryonic mes-

encephalic and diencephalic neurons appear under genetic control (ie, they are determined well before the appearance of any differences in reproductive steroid levels). Similarly, both the morphologic (eg, neuritic extension) and functional (eg, signal transduction) responses of cultured neurons and glia to reproductive steroids have been shown to display dramatic sexual dimorphisms despite exposure to identical levels of steroid,[17,51,52] ie, the dimorphic response cannot be attributed to differences in the steroid milieu of males and females. Additionally, the existence of brain sexual dimorphisms often is not translatable into gender-related differences in behavior.[53,54] Still, the widespread dimorphisms in animals (as well as the demonstrations of sexual dimorphisms in brain structure and physiology in humans)[55] provide a basis for inferring the mechanisms underlying reported gender dimorphisms in depression and other psychiatric disorders, ie, differences in prevalence, phenomenology (including characteristic symptoms, age of onset, susceptibility to recurrence, and stress responsivity), and treatment response characteristics.

Sexual dimorphisms

Sexual dimorphisms in depression

Depression differs in women and men in a number of respects. Studies consistently demonstrate a twofold increased prevalence of depression in women compared with men,[56-59] and this increased prevalence has been observed in a variety of countries.[58] A two- to threefold increased prevalence of dysthymia and threefold increase in seasonal affective disorder in women has also been noted,[60,61] while bipolar illness is equiprevalent in men and women[56,62,63] (reviewed in reference 64).

Prepubertal depression prevalence rates are not higher in girls,[65,66] possibly reflecting ascertainment bias (depressed boys may be more likely to come to the attention of health care providers) or the possibility that prepubertal major depression is premonitory of bipolar illness.[67] With some exceptions, the age of onset[58,59,68-71] (but also see references 72 to 75), type of symptoms, severity, and likelihood of chronicity and recurrence[58,59,68,72,76-78] (but also see references 79 to 83) display few differences between men and women. Women are more likely to present with anxiety, atypical symptoms, or somatic symptoms[60,68,72,81,83,85] are more likely to report symptoms (particularly in self-ratings),[60,68,85] are more likely to report antecedent stressful events,[86,87] and manifest a more robust effect of stress on the likelihood of developing depression during adolescence.[88] Women also display increased comorbidity of anxiety and eating disorders,[73,89-91] thyroid disease,[92,93] and migraine headaches,[94] as well as lower lifetime prevalence of substance abuse and dependence.[72,73] Reported differences in treatment response characteristics in women compared with men include poor response to tricyclics[95-98] particularly in younger women,[96] superior response to selective serotonin reuptake inhibitors (SSRIs) or monoamine oxidase inhibitors (MAOIs),[99,100] and a greater likelihood of response to triiodothyronine (T3) augmentation.[93] The extent to which these differences reflect gender-related differences in pharmacokinetics[101-107] remains to be determined. Finally, while the prevalence of bipolar disorder is comparable in men and women, women are more likely to develop rapid cycling[64] and may be more susceptible to antidepressant-induced rapid cycling.[108]

It is one matter to identify sex-related differences in depression and quite another to interpret their meaning. Specifically, one cannot infer that the observed differences are a product or a reflection of sex-specific biology. Sex-related differences in the prevalence of depression, for example, could occur consequent to increased number and severity of stressors experienced by women (eg, greater demands to manage both home and vocational responsibilities; societal encouragement of conciliatory behavior and discouragement of expression of anger; a lack of social empowerment) or to social stigmatization of endorsement of depressive symptoms in men. Nonetheless, the potential for sex-dependent biology to play a significant role in affective and cognitive disorders is suggested by the following (described below): (i) sexual dimorphisms in brain structure and physiology have been identified in humans; (ii) reproductive steroids regulate brain function in humans in vivo; and (iii) reproductive steroids play a role in the precipitation and treatment of mood disorders that are linked to periods of reproductive endocrine change.

Brain sexual dimorphisms in humans

While a biological basis for sex-dependent differences in the susceptibility to or expression of depression has not been demonstrated, structural and functional imaging studies have identified a variety of sex-differences in

the human brain, including the following: (i) functional organization of the brain, with brain activation response to rhyming task lateralized in men but not women[109]; (ii) gender-specific decreases in regional brain volume (caudate in men and globus pallidus, putamen in women) during development[110]; (iii) increased neuronal density in the temporal cortex in women[31]; (iv) greater interhemispheric coordinated activation of brain regions in women[111]; (v) larger volume hypothalamic nucleus (interstitial nucleus of the anterior hypthalamus–3 [INAH–3]) in men[112]; (vi) differences in both resting blood flow and the activation pattern accompanying self-induced mood change[113]; (vii) decreased serotonin receptor 5-HT_2 binding in the frontal, parietal, temporal, and cingulate cortices in women[114]; (viii) differences in whole brain serotonin synthesis (interpreted as decreased in women but possibly increased if corrected for plasma free tryptophan levels[115]); (ix) higher and more symmetric cerebral blood flow in women[116-120]; (x) greater asymmetry in the planum temporale in men[121]; and (xi) greater brain glucose metabolism (19%) in women.[122,123] Data from several studies employing similar technologies suggest that reproductive steroids may mediate some of the observed dimorphisms.

Regulation of brain physiology by reproductive steroids

Berman et al[124] demonstrated that the normal pattern of cognitive task–activated cerebral blood flow was eliminated by induced hypogonadism and restored by replacement with estradiol or progesterone, findings supported by Shaywitz et al,[125] who demonstrated estrogen enhancement of cognitive task–stimulated brain regional activation (functional magnetic resonance imaging [fMRI]) in postmenopausal women. Wong et al[126] demonstrated in a small number of subjects that dopamine receptor density in the caudate (measured by positron emission tomography [PET]) varied as a function of the menstrual cycle (lower in the follicular phase). Further, in two recent studies using paired-pulse transcranial magnetic stimulation, Smith et al[127,128] showed that cortical facilitation was enhanced in the late follicular phase, while cortical inhibition was enhanced during the luteal phase, consistent with putative central excitatory effects of estradiol and inhibitory effects of progesterone metabolites.

Despite gender-related and reproductive steroid–related differences in brain physiology, it is the investigation of mood disorders linked to reproductive endocrine change that offers the greatest potential insight into the role of reproductive steroids in the regulation and dysregulation of affect.

Reproductive endocrine–related mood disorders

Premenstrual syndrome

While Frank is credited with the first description of "premenstrual tension" in 1931, reports of mood and behavioral disturbances confined to the luteal phase of the menstrual cycle appeared earlier in the medical literature of the 19th century. For example, in 1847, Dr Ernst G. Von Feuchtersleben stated that "the menses in sensitive women is almost always attended by mental uneasiness, irritability or sadness."[129] As the symptoms of PMS occurred in a menstrual cycle phase–specific fashion (ie, only in the luteal phase), it was presumed that abnormalities in the hormonal constituents of the menstrual cycle (eg, estradiol, progesterone) must underlie PMS. Despite the appeal of this hormone excess or deficiency hypothesis, however, early studies of the putative hormonal etiologies of PMS were inconsistent in their conclusions. A major source of study inconsistency was identified in the 1980s,[130] namely that samples of women with PMS were selected (diagnosed) with highly unreliable techniques (ie, unconfirmed history). Without prospective demonstration of luteal phase–restricted symptom expression, samples selected were certain to contain a large number of false positives, thus rendering the data obtained ungeneralizable to the population with PMS.[131] This requirement for prospective confirmation of luteal phase symptomatology was ultimately incorporated into diagnostic criteria for PMS[132] and late luteal phase dysphoric disorder (LLPDD)/premenstrual dysphoric disorder (PMDD).[133] While the use of these diagnostic criteria/guidelines has permitted greater homogeneity of samples across studies—a requirement for comparison and generalization of results obtained—data subsequently generated have provided little if any evidence for hormonal excess or deficiency as etiologically relevant in PMS. Indeed, more recent studies have, if anything, largely preserved the formerly observed inconsistency. For example, Wang et al[134] observed increased estradiol and decreased progesterone levels in women with PMS, Redei and Freeman[135] reported nonsignificant

increases in both estradiol and progesterone, while Facchinetti et al[136] found no differences from controls in integrated progesterone levels. Results from studies of androgen levels have been similarly inconsistent, demonstrating both normal and decreased testosterone levels[137-139] and elevated and decreased free testosterone levels.[138,139] In conclusion, there is no consistent or convincing evidence that PMS is characterized by abnormal circulating plasma levels of gonadal steroids or gonadotropins or by hypothalamic-pituitary-ovarian axis dysfunction. Several studies do, however, suggest that levels of estradiol, progesterone, or neurosteroids (eg, pregnenolone sulfate) may be correlated with symptom severity in women with PMS.[134,140,141] (See references 142 and 143 for summaries of hormonal studies of PMS.)
If PMS is not due to a deficiency or excess of reproductive steroids (or of any other hormone studied to date), do these steroids play any role at all in the precipitation of the syndrome? We attempted to answer this question by posing four questions.

Is the luteal phase necessary for the appearance of PMS?

If there was no obvious abnormality in the activity of the reproductive axis, was PMS in fact dependent on the menstrual cycle for its expression, or could it be dissociated from the luteal phase? We blinded women to their position in the menstrual cycle by administering the progesterone receptor antagonist RU-486 (which both precipitates menses and ends corpus luteum activity), alone or with human chorionic gonadotropin (hCG) (which preserves corpus luteum activity).[144] Thus, after receiving the RU-486 (6 days after the LH surge), subjects did not know whether they were in the follicular phase of the next cycle (RU-486 alone) or in the preserved luteal phase of the initial cycle (RU-486 + hCG). Subjects in all three groups (a placebo-only group was included) experienced highly comparable symptoms that were significantly greater than those seen in the follicular phase; ie, women receiving RU-486 alone developed characteristic symptoms of PMS in the experimentally produced follicular phase of the next cycle. PMS, therefore, was not dependent on reproductive endocrine changes occurring in the mid-late luteal phase, as we were able to eliminate those changes without influencing subsequent symptom development. This left open the question of whether events occurring earlier than the mid-late luteal phase might, nonetheless, be influencing subsequent symptom development.

If you suppress ovarian activity, can you prevent the symptoms of PMS?

As the RU-486 study eliminated only the mid-late luteal phase, PMS symptoms might have appeared consequent to reproductive endocrine events occurring earlier in the menstrual cycle. To test this possibility, we performed "medical oophorectomies" by administering the gonadotropin-releasing hormone (GnRH) agonist leuprolide acetate (3.75 mg) in a placebo-controlled parallel-design study in 20 women with PMS. Leuprolide but not placebo was highly effective in eliminating both symptom severity and cyclicity (10/18 women responded to leuprolide and 0/10 responded to placebo).[145] This confirmed similar observations by Bancroft et al[146] and Mortola et al,[147] and suggested that PMS was indeed dependent upon ovarian steroid production.

In those in whom ovarian suppression effectively prevents the expression of PMS, will exogenous administration of gonadal steroids (either estrogen or progesterone) precipitate the return of characteristic symptoms?

Eighteen women whose PMS symptoms were significantly attenuated or eliminated by leuprolide-induced ovarian suppression were then continued on leuprolide and received in addition (in a double-blind, crossover fashion) estradiol (4 weeks followed by a fifth week in combination with progesterone to promote endometrial shedding) and progesterone (4 weeks). Five of these women received an additional 1 month of placebo "addback" in order to control for patients' expectations, specifically the recognition that they were taking something new. Finally, the same regimen of leuprolide-induced hypogonadism followed by sequential hormone replacement was performed in 15 control women, in whom the absence of menstrual cycle–related mood disturbances was confirmed with longitudinal ratings prior to study entry. The women with PMS whose symptoms were successfully eliminated (or attenuated) by leuprolide-induced hypogonadism experienced significant return of symptoms on either estradiol or progesterone, but not on placebo. Characteristically, symptoms returned within 2 weeks of initiating hormone replace-

ment and remitted by the fourth week of administration. In the control women lacking a history of PMS, however, neither the hypogonadal nor the hormone replacement conditions were associated with any perturbation of mood.[145] Consistent with the findings from our basal hormone studies,[148] it appeared that PMS represents an abnormal response to normal hormone changes or levels rather than a "normal" response to a hormonal abnormality. This study, then, raised a fundamental question: Why do similar changes in or levels of gonadal steroids trigger mood deterioration in women with PMS, while showing no apparent effect on mood in women lacking this history?

What are the potential mechanisms underlying the increased vulnerability to gonadal steroid–triggered mood changes in women with PMS?

While mood disorders may be seen in association with the pathological function of certain endocrine organs (eg, adrenal, thyroid), mood disturbances precipitated by gonadal steroids in PMS appear in the context of normal ovarian function. There are several possible means by which otherwise normal steroid signals might elicit a change in behavioral state.

Altered set points for relevant neural systems (ie, a means of conferring vulnerability)

The serotonin systems are appealing candidates for conferring vulnerability to gonadal steroid–precipitated mood changes[149]: gonadal steroids and serotonin display numerous reciprocal regulatory effects in the central nervous system (CNS); aggression against intruders by female rats (resident intruder model) varies with the estrous cycle, is ovarian steroid dependent, and is prevented by serotonin agonist antidepressants[150]; serotonin has a role in behaviors (eg, appetite, impulsivity, mood, sleep, and sexual interest) that vary with the menstrual cycle in PMS; blunted endocrine responses to serotonergic agonists (eg, L-tryptophan, *meta*-chlorophenylpiperazine) have been described in PMS (although not confined to the luteal phase)[151,152]; serotonergic agonists are efficacious in the treatment of PMS,[153] and the therapeutic efficacy of serotonin agonists can be reversed by tryptophan depletion[154] or serotonin receptor blockade.[155] While alterations in serotonin function are clearly relevant to the successful treatment of PMS symptoms, it remains unclear whether alterations in serotonin function underlie the predisposition to experience PMS. Future studies will await the development of receptor subtype specific agonists/antagonists and access to subtype-specific imaging ligands.

Polymorphisms in gonadal steroid signaling pathway proteins or in systems regulated by gonadal steroids

PMS offers an ideal opportunity to identify genetic contributions to the vulnerability for affective disturbance, since the offending stimuli (steroid triggers) are known. Several polymorphisms in gonadal steroid receptors have been shown to alter receptor transcriptional efficacy (eg, CAG repeat in exon 1 of the androgen receptor; progins insertion in intron 7 of the progesterone receptor) and to be associated with differential illness risk (ie, prostate cancer or breast cancer).[156-159] Additionally, the susceptibility to the disruptive effects of estradiol on reproductive development differs enormously (up to 100-fold) between mouse strains, with the genotype contributing more to the variance than the dose of estradiol employed.[160] There is precedent, then, for inferring that polymorphisms in the gonadal steroid–signaling pathway or in gonadal steroid–regulated genes may alter the nature or strength of the steroid signal as well as phenotype. In genetic studies that we have performed to date in 125 women with PMS and 280 controls (C. Roca and B. Harlow, unpublished data), no differences were observed in the frequencies of the following polymorphisms: PvuII, XbaI, and TA repeat (estrogen receptor α); CAG repeat (androgen receptor); progins, CA repeat (progesterone receptor); T102C, His-Tyr (serotonin 2A receptor); Cys-Ser (serotonin 2C receptor). A significant difference has been identified, however, for the SLC6A4 promoter polymorphism of the serotonin transporter, with a higher frequency of the L (long) allele (associated with increased transport and increased response to SSRIs)[161,162] in the women with PMS (C. Roca et al, unpublished data). This difference appears to reflect a lower than predicted frequency in the controls as well as a higher frequency in patients, suggesting that women with documented absence of any menstrual cycle–related mood symptoms (approximately 10% of the Harlow sample) may be protected from the development of symptoms and hence may be at least as informative as illness probands in providing clues to the genetic determinants of susceptibility.

Altered metabolism of gonadal steroids

The neurosteroid metabolites of progesterone (and androgens) are of considerable interest as possible mediators of the behavioral effects of gonadal steroids. Supportive observations are as follows: (i) the ring A–reduced metabolites of progesterone, allopregnanolone and pregnanolone, are allosteric modulators of the $GABA_A$ receptor/chloride ionophore[21]; (ii) withdrawal of progesterone in rats produces anxiety and insensitivity to benzodiazepines due to withdrawal of allopregnanolone, with consequent induction of $GABA_A$ alpha-4 subunit levels and inhibition of GABA currents[163,164]; (iii) decreased plasma allopregnanolone levels are seen in major depressive disorder and in depression associated with alcohol withdrawal, with an increase in levels seen in plasma and cerebrospinal fluid (CSF) following successful antidepressant treatment[165-167]; (iv) allopregnanolone displays anxiolytic effects in several animal anxiety models[168] and may be involved in the stress response[169]; (v) antidepressants may promote the reductive activity of one of the neurosteroid synthetic enzymes (3α-HSOR), thus favoring the formation of allopregnanolone.[170] While we previously reported no differences in luteal phase allopregnanolone and pregnanolone levels in women with PMS compared with controls,[171] in an experimental model of postpartum depression (PPD), we observed a highly significant inverse correlation (r=0.92) between the change in allopregnanolone levels from weeks 6 to 8 of hormone addback and Beck depression ratings at week 8 of addback (see below).[172] This correlation reflected the high depression ratings in those women with a past history of PPD, whose allopregnanolone levels dropped or failed to increase during the last 2 weeks of high dose addback. These findings suggest that differences in the activity of the synthetic (or metabolic) enzymes for neurosteroids may translate into phenotypic differences.

At present, we do not understand the basis for the differential sensitivity seen in women with PMS: what permits levels or changes in reproductive steroids that are without effect on mood in women without a history of PMS to destabilize mood in those with a history of PMS? Nonetheless, the simultaneous requirement for a trigger (reproductive steroids) and an underlying vulnerability to mood state destabilization in PMS provides a model for thinking about affective disorders in general as well as other mood disorders (eg, postpartum depression) specifically linked to periods of reproductive endocrine change.

Postpartum depression

A reproductive endocrine–related mood disorder that is phenomenologically similar to major depression is PPD, the most prominent symptoms of which are sleep disturbance, excessive fatigue, sadness and anhedonia, excessive guilt or self blame, psychomotor disturbance, and suicidal ideation.[173-175] It does not appear that there is anything phenomenologically unique about the depression that occurs postpartum; rather, once again, it is the timing of the syndrome that makes it distinctive, in this case following delivery.

However, variability in the definition of the interval during which PPD can develop (2 weeks to 3 months postpartum) in part accounts for the variable estimates of the incidence and prevalence of PPD. Prevalence rates for PPD vary between 8.3% and 14.9%.[176-181] While an increased prevalence of depression postpartum has not been clearly demonstrated (due, in part, again to varying intervals examined and a paucity of adequate control groups), it does appear that the relative risk of depression increases during the first few months postpartum.[178,182-184]

While a variety of factors have been associated with the development of PPD, including personal or family history of psychiatric illness, marital disharmony, lack of confiding relationships, and number of life events in the previous year,[185-187] two are of particular interest. First, while some but not all studies show a prior history of affective illness as a risk factor for subsequent PPD,[188-191] women with PPD as their first depressive episode appear both less likely to experience a nonpuerperal depression and more likely to experience a subsequent PPD than women with nonpuerperal episodes.[192] Second, recent studies suggest that depressive symptoms during pregnancy may be associated with the development of PPD.[184,189-191,193-195] Any hypothetical role of reproductive steroids in PPD must account for the increase in depressive symptoms during pregnancy.

Studies have examined the relationship between PPD and reproductive steroids by measuring steroid levels (particularly estradiol and progesterone) or changes in levels during pregnancy and the postpartum. The results of these studies in general fail to show any consistent differences between women with PPD and controls.[196] Similarly, while thyroid dysfunction may contribute to postpartum mood dysregulation in a small group of women, it does not appear relevant for the majority of

women with PPD. PPD, then, cannot be thought of as a simple hormonal excess or deficiency state. If there is no reproductive endocrine abnormality in women with PPD, and symptoms, at least in some cases, develop during pregnancy, could PPD represent an altered sensitivity to reproductive steroids in a subgroup of women? Supportive evidence for this role of differential sensitivity is drawn from two indirect sources. First, vulnerability to PPD appears to associate with vulnerability to other reproductive endocrine–related mood disorders. Several studies,[197,198] for example, report marked elevations in the prevalence of a history of PPD (up to 68%) in women with PMS, and high postpartum depressive scores have been associated with a history of PMS.[199,200] Second, the relevance of reproductive steroids is suggested by recent reports of the efficacy of hormonal treatments of PPD. These reports suggest the acute[201] and prophylactic[202] antidepressant effects of estradiol in women with PPD, with the recurrence rate in the latter study reduced to 9% from an anticipated rate of 35% to 60%.

Direct evidence in support of the role of reproductive steroids in the development of PPD comes from a study in which a scaled-down form of pregnancy and parturition was created in euthymic women with and without a history of PPD. Use of this model permitted examination of the role of reproductive steroids in postpartum mood symptoms without many of the factors that confound efforts to study PPD, including dramatic concurrent changes in other endocrine axes (eg, hypothalamopituitary-adrenal [HPA] axis), obstetrical pain and complications, varying levels of social support, and stress secondary to childbirth and motherhood. The GnRH agonist leuprolide acetate was used to suppress ovarian steroid production and create a stable hypogonadal baseline, following which supraphysiologic doses of estradiol and progesterone were administered for 2 months and then abruptly (and blindly) withdrawn. This methodology replicated (albeit on a smaller scale) both the elevated reproductive steroid levels seen during pregnancy and the precipitous decline in levels at parturition. Five of the eight women with a history of PPD, and none of the controls, developed significant mood symptoms during both hormone addback and withdrawal, findings consistent with observations that the incidence of depressive symptoms is increased during both the last trimester and postpartum. This study suggests a direct role for estradiol or progesterone or both in PPD and further demonstrates that women with a history of PPD are differentially sensitive to the mood-destabilizing effects of marked changes in levels of reproductive steroids.

Conclusions

The differential sensitivity to gonadal steroids seen in women with histories of PMS and PPD emphasizes that the response to a biological signal cannot be inferred without an understanding of the context in which the signal occurs. This context includes current physiological and external environments, prior experience, past history of exposure to the stimulus, and genetic makeup. With the mapping of the human genome, this last contextual determinant becomes of great practical interest as a potential explanation for differential response to steroids.

While genetic polymorphisms clearly influence the transcriptional, physiological, and behavioral responses to activated steroid receptors, it seems equally clear that genetic factors will contribute to, but not solely provide, the explanation for the differential sensitivity seen in women with PMS and PPD. Indeed, even where genetic strain differences are apparent in behavioral sensitivity to reproductive steroids, not all strain members demonstrate the observed steroid-stimulated behavior.[150] Further, the observed alterations in reproductive steroid–sensitive neurocircuitry, reproductive steroid–activated gene expression, and adult behavior following differential exposure to perinatal steroids[23,203,204] caution us that gene–environment interactions may yield markedly different phenotypic expressions of the same genotype. The variable influences on behavior, then, are not likely to reduce to simple, unitary explanations for the susceptibility to depression. Nonetheless, by recognizing that a biological stimulus may trigger an affective state change (operate as an affective trigger) only in a specific context of susceptibility, we are in a much better position to meaningfully explore and uncover the pathophysiology of depression. By illuminating the mechanisms underlying the differential sensitivity to reproductive steroids exemplified by women with PMS and PPD, we will significantly advance our understanding of the neurobiology of affective illness. ❑

Esteroides gonadales, cerebro y conducta: papel del contexto

La naturaleza y extensión de la influencia del sexo y la función reproductora en el ánimo ha sido objeto por siglos de especulación y controversia. En los últimos 50 años; sin embargo, se ha demostrado cada vez con mayor claridad que no es sólo el cerebro el blanco principal de las hormonas esteroidales de la reproducción, sino que adicionalmente, las hormonas esteroidales – como neurorreguladores- dan origen a un contexto que influye en una amplia gama de actividades cerebrales. Por ejemplo, las acciones neurales y las conductas resultantes son marcadamente diferentes en presencia o ausencia de esteroides gonadales. A su vez, las acciones de los esteroides gonadales son por sí mismas dependientes del contexto. De este modo, aun cuando se pueda demostrar que los esteroides gonadales provocan trastornos del ánimo, esta provocación occurre con niveles hormonales normales (en oposición a los trastornos del ánimo que acompañan a las endocrinopatías) y los trastornos del ánimo aparecen sólo en un subgrupo de individuos sensibles. La especificidad del contexto y la diferente susceptibilidad para la disregulación afectiva que se observa en mujeres con trastornos del ánimo relacionados con el ciclo reproductor endocrino, son sin lugar a dudas importantes características que subyacen a una amplia gama de trastornos psiquiátricos en los que los factores gatillo aun no han sido identificados. En consecuencia, los trastornos del ánimo relacionados con el ciclo reproductor endocrino ofrecen una esperanza sin igual para la identificación de aquellas variables del contexto que permiten que estímulos biológicos se transformen específicamente en depresión en individuos que están en riesgo.

Stéroïdes gonadiques, cerveau et comportement : rôle du contexte

La nature et l'importance de l'influence des fonctions sexuelles et reproductives sur l'humeur ont été l'objet de discussion et de controverse depuis des siècles. Cependant, depuis les 50 dernières années, il est devenu de plus en plus évident que non seulement le cerveau est une cible majeure des hormones stéroïdiennes de la reproduction, mais qu'en plus les hormones stéroïdiennes, en tant que neurorégulateurs, créent un contexte qui influe sur une large gamme d'activités cérébrales, c'est-à-dire, qu'en fonction de la présence ou de l'absence des stéroïdes gonadiques, les effets neurologiques et les comportements en résultant sont très différents. De plus, les effets des hormones stéroïdiennes gonadiques dépendent eux-mêmes du contexte. Par conséquent, même quand il est possible de démontrer que les hormones stéroïdiennes provoquent des troubles de l'humeur, ces derniers apparaissent pour des concentrations normales d'hormones stéroïdiennes (par opposition aux troubles de l'humeur qui accompagnent les endocrinopathies), et seulement dans un sous-groupe d'individus sensibles. La spécificité du contexte et la différence de sensibilité individuelle aux désordres affectifs, décrites chez des femmes ayant des troubles de l'humeur liés aux hormones de la reproduction, sont de toute évidence des particularités importantes sous-jacentes d'un large échantillon de troubles psychiatriques pour lesquels les facteurs déclenchants n'ont pas encore été identifiés. Par conséquent, les troubles de l'humeur liés aux hormones de la reproduction offrent une possibilité sans précédent d'identifier les variables de ce contexte qui permettent aux stimuli biologiques de se traduire de façon différente en dépression chez les individus à risque.

REFERENCES

1. Dorfman RI, Shipley RA. *Androgens: Biochemistry, Physiology, and Clinical Significance*. New York, NY: John Wiley & Sons; 1956.
2. Brown-Séquard CE. The effects produced on man by subcutaneous injections of a liquid obtained from the testicles of animals. *Lancet*. 1889;2:105-107.
3. Easterbrook CC. Organo-therapeutics in mental diseases. *BMJ*. 1900;2:813-823.
4. Brown-Séquard CE. On a new therapeutic method consisting in the use of organic liquids extracted from glands and other organs. *BMJ*. 1893;1:1145-1147, 1212-1214.
5. Wilson JD. Charles-Edouard Brown-Séquard and the centennial of endocrinology. *J Clin Endocrinol Metab.* 1990;71:1403-1409.
6. Jensen EV, Jacobson HI. Basic guides to the mechanism of estrogen action. *Recent Prog Horm Res.* 1962;18:387-414.
7. Toft D, Gorski J. A receptor molecule for estrogens: isolation from the rat uterus and preliminary characterization. *Proc Natl Acad Sci U S A*. 1966;55:1574-1581.
8. McEwen BS. Basic neurobiology of ovarian steroids: clinical implications. *Dialogues Clin Neurosci.* 2002;4:163-175.
9. Halachmi S, Marden E, Martin G, MacKay H, Abbondanza C, Brown M. Estrogen receptor-associated proteins: possible mediators of hormone-induced transcription. *Science*. 1994;264:1455-1458.

10. Uht RM, Anderson CM, Webb P, Kushner PJ. Transcriptional activities of estrogen and glucocorticoid receptors are functionally integrated at the AP-1 response element. *Endocrinology*. 1997;138:2900-2908.
11. Smith CL, Nauaz Z, O'Malley BW. Co-activator and co-repressor regulation of the agonist/antagonist activity of the mixed antiestrogen, 4-hydroxytamoxifen. *Mol Endocrinol*. 1997;11:657-666.
12. Jackson TA, Richer JK, Bain DL, Takimoto GS, Tung L, Horwitz KB. The partial agonist activity of antagonist-occupied steroid receptors is controlled by a novel hinge domain-binding co-activator L7/SPA and the corepressors N-COR or SMRT. *Mol Endocrinol*. 1997;11:693-705.
13. Kuiper GGJM, Shughrue PJ, Merchenthaler I, Gustafsson JA. The estrogen receptor β subtype: a novel mediator of estrogen action in neuroendocrine systems. *Front Neuroendocrinol*. 1998;19:253-286.
14. Moore JT, McKee DD, Slentz-Kesler K, et al. Cloning and characterization of human estrogen receptor β isoforms. *Biochem Biophys Res Commun*. 1998;247:75-78.
15. Shughrue PJ, Lane MV, Merchenthaler I. Comparative distribution of estrogen receptor-α and -β mRNA in the rat central nervous system. *J Comp Neurol*. 1997;388:507-525.
16. Paech K, Webb P, Kuiper GGJM, et al. Differential ligand activation of estrogen receptors ERα and ERβ at AP1 sites. *Science*. 1997;277:1508-1510.
17. Zhang L, Li B, Zhao W, et al. Sex-related differences in MAPKs activation in rat astrocytes: effects of estrogen on cell death. *Mol Brain Res*. 2002. In press.
18. Grino PB, Griffin JE, Wilson JD. Testosterone at high concentrations interacts with the human androgen receptor similarly to dihydrotestosterone. *Endocrinology*. 1990;126:1165-1172.
19. Naftolin F, Garcia-Segura LM, Keefe D, Leranth C, MacLusky NJ, Brawer JR. Estrogen effects on the synaptology and neural membranes of the rat hypothalamic arcuate nucleus. *Biol Reprod*. 1990;42:21-28.
20. Mahendroo MS, Cala KM, Landrum DP, Russell DW. Fetal death in mice lacking 5α-reductase type 1 caused by estrogen excess. *Mol Endocrinol*. 1997;11:917-927.
21. Majewska MD, Harrison NL, Schwartz RD, Barker JL, Paul SM. Steroid hormone metabolites are barbiturate-like modulators of the GABA receptor. *Science*. 1986;232:1004-1007.
22. Poletti A, Celotti F, Maggi R, Melcangi RC, Martini L, Negri-Cesi P. Aspects of hormonal steroid metabolism in the nervous system. In: Baulieu EE, Robel P, Schumacher M, eds. *Contemporary Endocrinology: Neurosteroids. A New Regulatory Function in the Nervous System*, Totowa, NJ: Humana Press Inc; 1999:97-123.
23. Phoenix CH, Goy RW, Gerall AA, Young WC. Organizing action of prenatally administered testosterone propionate on the tissues mediating mating behavior in the female guinea pig. *Endocrinology*. 1959;65:369-382.
24. Gorski RA, Gordon JH, Shryne JE, Southam AM. Evidence for a morphological sex difference within the medial preoptic area of the rat brain. *Brain Res*. 1978;148:333-346.
25. Toran-Allerand CD. Developmental interactions of estrogens with the neurotrophins and their receptors. In: Micevych P, Hammer RP, eds. *Neurobiological Effects of Sex Steroid Hormones*. Cambridge, UK: Cambridge University Press; 1994:391-411.
26. Miranda P, Williams CL, Einstein G. Granule cells in aging rats are sexually dimorphic in their response to estradiol. *J Neurosci*. 1999;19:3316-3325.
27. Watters JJ, Campbell JS, Cunningham MJ, Krebs EG, Dorsa DM. Rapid membrane effects of steroids in neuroblastoma cells: effects of estrogen on mitogen-activated protein kinase signalling cascade and c-fos immediate early gene transcription. *Endocrinology*. 1997;138:4030-4033.
28. Garcia-Segura LM, Cardona-Gomez P, Naftolin F, Chowen JA. Estradiol upregulates Bcl-2 expression in adult brain neurons. *Neuroendocrinology*. 1998;9:593-597.
29. Gouras GK, Xu H, Gross RS, Greenfield JP, Hai B, Wang R, Greengard P. Testosterone reduces neuronal secretion of Alzheimer's beta-amyloid peptides. *Proc Natl Acad Sci U S A*. 2000;97:1202-1205.
30. Zhang L, Li BS, Ma W, et al. Dehydroepiandrosterone (DHEA) and its sulfated derivative (DHEAS) regulate apoptosis during neurogenesis by triggering the Akt signaling pathway in opposing ways. *Mol Brain Res*. 2002;98:58-66.
31. Witelson SA. Neural sexual mosaicism: sexual differentiation of the human temporo-parietal region for functional asymmetry. *Psychoneuroendocrinology*. 1991;16:131-153.
32. Ongur D, Drevets WC, Price JL. Glial reduction in the subgenual prefrontal cortex in mood disorders. *Proc Natl Acad Sci U S A*. 1998;95:13290-13295.
33. Rajkowska G, Miguel-Hidalgo JJ, Wei J, et al. Morphometric evidence for neuronal and glial prefrontal cell pathology in major depression. *Biol Psychiatry*. 1999;45:1085-1098.
34. Rajkowska G. Postmortem studies in mood disorder indicate altered numbers of neurons and glial cells. *Biol Psychiatry*. 2000;48:766-777.
35. Post RM, Rubinow DR, Ballenger JC. Conditioning and sensitisation in the longitudinal course of affective illness. *Br J Psychiatry*. 1986;149:191-201.
36. Antelman SM, Caggiula AR, Kocan D, et al. One experience with "lower" or "higher" intensity stressors, respectively enhances or diminishes responsiveness to haloperidol weeks later: implications for understanding drug variability. *Brain Res*. 1991;566:276-283.
37. Weiss SR, Post RM, Pert A, Woodward R, Murman D. Context-dependent cocaine sensitization: differential effect of haloperidol on development versus expression. *Pharmacol Biochem Behav*. 1989;34:655-661.
38. Liu D, Diorio J, Tannenbaum B, et al. Maternal care, hippocampal glucocorticoid receptors, and hypothalamic-pituitary-adrenal responses to stress. *Science*. 1997;277:1659-1662.
39. Levine S. Infantile experience and resistance to physiological stress. *Science*. 1975;126:405-406.
40. Francis D, Diorio J, Liu D, Meaney MJ. Nongenomic transmission across generations of maternal behavior and stress responses in the rat. *Science*. 1999;286:1155-1158.
41. Ward IL, Weisz J. Differential effects of maternal stress on circulating levels of corticosterone, progesterone, and testosterone in male and female rat fetuses and their mothers. *Endocrinology*. 1984;114:1635-1644.
42. Ward IL, Weisz J. Maternal stress alters plasma testosterone in fetal males. *Science*. 1980;207:328-329.
43. Ward IL, Stehm KE. Prenatal stress feminizes juvenile play patterns in male rats. *Physiol Behav*. 1991;50:601-605.
44. Moore CL, Dou H, Juraska JM. Maternal stimulation affects the number of motor neurons in a sexually dimorphic nucleus of the lumbar spinal cord. *Brain Res*. 1992;572:52-56.
45. Schechter D, Gandelman R. Inter-male aggression in mice: influence of gonadectomy and prior fighting experience. *Aggress Behav*. 1981;7:187-193.
46. Pfaff DW. Morphological changes in the brains of adult male rats after neonatal castration. *J Endocrinol*. 1966;36:415-416.
47. Raisman G, Field PM. Sexual dimorphism in the preoptic area of the rat. *Science*. 1971;173:731-733.
48. Gorski RA. Sexual differentiation of the endocrine brain and its control. In: Motta M, ed. *Brain Endocrinology*. New York, NY: Raven Press; 1991:71-104.
49. Pilgrim C, Hutchison JB. Developmental regulation of sex differences in the brain: can the role of gonadal steroids be redefined? *Neuroscience*. 1994;60:843-855.
50. Reisert L, Pilgrim C. Sexual differentiation of monoaminergic neurons—genetic or epigenetic? *Trends Neurosci*. 1991;14:468-473.
51. McEwen BS, Biegon A, Fischette CT, Luine VN, Parsons B, Rainbow TC. Sex differences in programming of responses to estradiol in the brain. In: Serio M, Motta, M, Zanisi M, Martini L, eds. *Sexual Differentiation: Basic and Clinical Aspects.* New York, NY: Raven Press; 1984:93-98.
52. Luine VN. Estradiol increases choline acetyltransferase activity in specific basal forebrain nuclei and projection areas of female rats. *Exp Neurol*. 1985;89:484-490.
53. Arendash GW, Gorski RA. Effects of discrete lesions of the sexually dimorphic nucleus of the preoptic area or other medial preoptic regions on the sexual behavior of male rats. *Brain Res Bull.* 1983;10:147-150.
54. De Vries GJ, Boyle PA. Double duty for sex differences in the brain. *Behav Brain Res*. 1998;92:205-213.
55. Rubinow DR, Schmidt PJ, Roca CA. Hormonal and gender influences on mood regulation. In: Nemeroff CB, ed. *Neuropsychopharmacology: the Fifth Generation of Progress*, Baltimore, Md: Lippincott Williams & Williams. 2002. In press.
56. Psychiatric Disorders in America: the Epidemiologic Catchment Area Study. New York, NY: The Free Press; 1991.
57. Weissman MM, Klerman GL. Sex differences in the epidemiology of depression. *Arch Gen Psychiatry*. 1977;34:98-111.

58. Weissman MM, Bland R, Joyce PR, Newman S, Wells JE, Witchen HU. Sex differences in rates of depression: cross-national perspectives. *J Affect Disord*. 1993;29:77-84.
59. Kessler RC, McGonagle KA, Swartz M, Blazer DG, Nelson CB. Sex and depression in the National Comorbidity Survey I: lifetime prevalence, chronicity and recurrence. *J Affect Disord*. 1993;29:85-96.
60. Leibenluft E, Hardin TA, Rosenthal NE. Gender differences in seasonal affective disorder. *Depression*. 1995;3:13-19.
61. American Psychiatric Association. *Diagnostic and Statistical Manual of Mental Disorders*. 4th ed. Washington, DC: American Psychiatric Association; 1994.
62. Weissman MM, Klerman GL. Sex differences and the epidemiology of depression. In: Guggenheim FG, Nadelson CC, eds. *Major Psychiatric Disorders: Overviews and Selected Readings*. New York, NY: Elsevier Science Publishing; 1982:95-114.
63. Weissman MM, Klerman GL. Gender and depression. *Trends Neurosci*. 1985;8:416-420.
64. Leibenluft E. Women with bipolar illness: clinical and research issues. *Am J Psychiatry*. 1996;153:163-173.
65. Anderson JC, Williams S, McGee R, Silva PA. *DSM-III* disorders in preadolescent children: prevalence in a large sample from the general population. *Arch Gen Psychiatry*. 1987;44:69-77.
66. McGee R, Feehan M, Williams S, Anderson J. DSM-III disorders from age 11 to age 15 years. *J Am Acad Child Adolesc Psychiatry*. 1992;31:50-59.
67. Leibenluft E. Gender differences in major depressive disorder and bipolar disorder. *CNS Spectrums*. 1999;4:25-33.
68. Frank E, Carpenter LL, Kupfer DJ. Sex differences in recurrent depression: are there any that are significant? *Am J Psychiatry*. 1988;145:41-45.
69. Thase ME, Reynolds CF, Frank E, et al. Do depressed men and women respond similarly to cognitive-behavior therapy. *Am J Psychiatry*. 1994;151:500-505.
70. Burke KC, Burke JD, Regier DA, Rae DS. Age at onset of selected mental-disorders in five community populations. *Arch Gen Psychiatry*. 1990;47:511-518.
71. Winokur G, Tsuang MT, Crowe RR. The Iowa 500: affective disorder in relatives of manic and depressed patients. *Am J Psychiatry*. 1982;139:209-212.
72. Kornstein SG, Schatzberg AF, Yonkers KA, et al. Gender differences in presentation of chronic major depression. *Psychopharmacol Bull*. 1995;31:711-718.
73. Fava M, Abraham M, Alpert J, Nierenberg AA, Pava JA, Rosenbaum JF. Gender differences in axis I comorbidity among depressed outpatients. *J Affect Disord*. 1996;38:129-133.
74. Spicer CC, Hare EH, Slater E. Neurotic and psychotic forms of depressive illness; evidence from age-incidence in a national sample. *Br J Psychiatry*. 1973;123:535-541.
75. Nolen-Hoeksema S. Sex differences in unipolar depression: evidence and theory. *Psychol Bull*. 1987;101:259-282.
76. Kessler RC, McGonagle KA, Nelson CB, Hughes M, Swartz M, Blazer DG. Sex and depression in the National Comorbidity Survey II: cohort effects. *J Affect Disord*. 1994;30:15-26.
77. Simpson HB, Nee JC, Endicott J. First-episode major depression. *Arch Gen Psychiatry*. 1997;54:633-639.
78. Zlotnick C, Shea MT, Pilkonis PA, Elkin I, Ryan C. Gender, type of treatment, dysfunctional attitudes, social support, life events, and depressive symptoms over naturalistic follow-up. *Am J Psychiatry*. 1996;153:1021-1027.
79. Sargeant JK, Bruce ML, Florio LP, Weissman MM. Factors associated with 1-year outcome of major depression in the community. *Arch Gen Psychiatry*. 1990;47:519-526.
80. Aneshensel CS. The natural history of depressive symptoms. *Res Commun Ment Health*. 1985;5:45-74.
81. Ernst C, Angst J. The Zurich Study. XII. Sex differences in depression. Evidence from longitudinal epidemiological data. *Eur Arch Psychiatry Clin Neurosci*. 1992;241:222-230.
82. Keitner GI, Ryan CE, Miller IW, Kohn R, Epstein NB. 12-month outcome of patients with major depression and comorbid psychiatric or medical illness (compound depression). *Am J Psychiatry*. 1991;148:345-350.
83. Winokur G, Coryell W, Keller M, Endicott J, Akiskal H. A prospective follow-up of patients with bipolar and primary unipolar affective-disorder. *Arch Gen Psychiatry*. 1993;50:457-465.
84. Young MA, Fogg LF, Scheftner WA, Keller MB, Fawcett JA. Sex differences in the lifetime prevalence of depression: does varying the diagnostic criteria reduce the female/male ratio? *J Affect Disord*. 1990;18:187-192.
85. Angst J, Dobler-Mikola A. Do the diagnostic criteria determine the sex ratio in depression? *J Affect Disord*. 1984;7:189-198.
86. Bebbington PE, Brugha T, MacCarthy B, et al. The Camberwell Collaborative Depression Study, I: depressed probands—adversity and the form of depression. *Br J Psychiatry*. 1988;152:754-765.
87. Karp JF, Frank E. Combination therapy and the depressed woman. *Depression*. 1995;3:91-98.
88. Silberg J, Pickles A, Rutter M, et al. The influence of genetic factors and life stress on depression among adolescent girls. *Arch Gen Psychiatry*. 1999;56:225-232.
89. Blazer DG, Kessler RC, McGonagle KA, Swartz MS. The prevalence and distribution of major depression in a national community sample: the National Comorbidity Survey. *Am J Psychiatry*. 1994;151:979-986.
90. Regier DA, Burke JD, Burke KC. Comorbidity of affective and anxiety disorders in the NIMH Epidemiologic Catchment Area Program. In: Maser JD, Cloninger CR, eds. Comorbidity of Mood and Anxiety Disorders. Washington, DC: *American Psychiatric Press*; 1990:113-122.
91. Judd LL. When anxiety disorders are comorbid with major depression: social and clinical burden. 147th American Psychiatric Association Meeting. Philadelphia, Pa. 1994. Abstract.
92. Reus VI. Behavioral aspects of thyroid disease in women. *Psychiatr Clin North Am*. 1989;12:153-165.
93. Whybrow PC. Sex differences in thyroid axis function: relevance to affective disorder and its treatment. *Depression*. 1995;3:33-42.
94. Moldin SO, Scheftner WA, Rice JP, Nelson E, Knesevich MA, Akiskal H. Association between major depressive disorder and physical illness. *Psychol Med*. 1993;23:755-761.
95. Old Age Depression Interest Group. How long should the elderly take antidepressants? A double-blind placebo-controlled study of continuation/prophylaxis therapy with dothiepin. *Br J Psychiatry*. 1993;162:175-182.
96. Raskin A. Age-sex differences in response to antidepressant drugs. *J Nerv Ment Dis*. 1974;159:120-130.
97. Glassman AH, Perel JM, Shostak M, Kantor SJ, Fleiss JL. Clinical implications of imipramine plasma levels for depressive illness. *Arch Gen Psychiatry*. 1977;34:197-204.
98. Coppen A, Whybrow PC, Noguera R, Maggs R, Prange AJ, Jr. The comparative antidepressant value of L-tryptophan and imipramine with and without attempted potentiation by liothyronine. *Arch Gen Psychiatry*. 1972;26:234-241.
99. Davidson J, Pelton S. Forms of atypical depression and their response to antidepressant drugs. *Psychiatry Res*. 1986;17:87-95.
100. Kornstein SC, Schatzberg AF, Thase ME, et al. Gender differences in treatment response to sertraline versus imipramine in chronic depression. *Am J Psychiatry*. 2000;157:1445-1452.
101. Dawkins K, Potter WZ. Gender differences in pharmacokinetics and pharmacodynamics of psychotropics: focus on women. *Psychopharmacol Bull*. 1991;27:417-426.
102. Yonkers KA, Kando JC, Cole JO, Blumenthal S. Gender differences in pharmacokinetics and pharmacodynamics of psychotropic medication. *Am J Psychiatry*. 1992;149:587-595.
103. Moody JP, Tait AC, Todrick A. Plasma levels of imipramine and desmethylimipramine during therapy. *Br J Psychiatry*. 1967;113:183-193.
104. Preskorn SH, Mac DS. Plasma levels of amitriptyline: effects of age and sex. *J Clin Psychiatry*. 1985;46:276-277.
105. Gex-Fabry M, Balant-Gorgia AE, Balant LP, Garrone G. Clomipramine metabolism: model-based analysis of variability factors from drug monitoring data. *Clin Pharmacokinet*. 1990;19:241-255.
106. Greenblatt DJ, Friedman H, Burstein ES, et al. Trazodone kinetics: effect of age, gender, and obesity. *Clin Pharmacol Ther*. 1987;42:193-200.
107. Warrington SJ. Clinical implications of the pharmacology of sertraline. *Int Clin Psychopharmacol*. 1991;6:11-21.
108. Altshuler LL, Post RM, Leverich GS, Mikalauskas K, Rosoff A, Ackerman L. Antidepressant-induced mania and cycle acceleration: a controversy revisited. *Am J Psychiatry*. 1995;152:1130-1138.

109. Shaywitz BA, Shaywitz SE, Pugh KR, et al. Sex differences in the functional organization of the brain for language. *Nature*. 1995;373:607-609.
110. Giedd JN, Blumenthal J, Jeffries NO, et al. Brain development during childhood and adolescence: a longitudinal MRI study. *Nature*. 1999;2:861-863.
111. Azari NP, Pettigrew KD, Pietrini P, Murphy DG, Horwitz B, Schapiro MB. Sex-differences in patterns of hemispheric cerebral metabolism—a multiple-regression discriminant-analysis of positron emission tomographic data. *Int J Neurosci*. 1995;81:1-20.
112. Allen L, Hines M, Shryne J, Gorski R. Two sexually dimorphic cell groups in the human brain. *J Neurosci*. 1989;9:497-506.
113. George MS, Ketter TA, Parekh PI, Herscovitch P, Post RM. Gender differences in regional cerebral blood flow during transient self-induced sadness or happiness. *Biol Psychiatry*. 1996;40:859-871.
114. Biver F, Lotstra F, Monclus M, et al. Sex difference in $5HT_2$ receptor in the living human brain. *Neurosci Lett*. 1996;204:25-28.
115. Nishizawa S, Benkelfat C, Young SN, et al. Differences between males and females in rates of serotonin synthesis in human brain. *Proc Natl Acad Sci U S A*. 1997;94:5308-5313.
116. Rodriguez G, Warkentin S, Risberg J, Rosadini G. Sex differences in regional cerebral blood flow. *J Cereb Blood Flow Metab*. 1988;8:783-789.
117. Gur RC, Gur RE, Obrist WD, et al. Sex and handedness differences in cerebral blood flow during rest and during cognitive activity. *Science*. 1982;217:659-661.
118. Gur RC, Gur RE, Obrist WD, Skolnick BE, Reivich M. Age and regional cerebral blood flow at rest and during cognitive activity. *Arch Gen Psychiatry*. 1987;44:617-621.
119. Shaw T, Meyer JS, Mortel K, et al. Effects of normal aging, sex, and risk factors for stroke on regional cerebral blood flow (rCBF) in normal volunteers. In: Gotoh F, Hagai H, Tazaki Y, eds. *Cerebral Blood Flow and Metabolism*, Copenhagen, Denmark: Munksgaard; 1979.
120. Esposito G, Van Horn JD, Weinberger DR, Berman KF. Gender differences in cerebral blood flow as a function of cognitive state with PET. *J Nucl Med*. 1996;37:559-564.
121. Kulynych JJ, Vladar K, Jones DW, Weinberger DR. Gender differences in the normal lateralization of the supratemporal cortex: MRI surface-rendering morphometry of Heschl's gyrus and the planum temporale. *Cereb Cortex*. 1994;4:107-118.
122. Baxter LR, Jr, Mazziotta JC, Phelps ME, Selin CE, Guze BH, Fairbanks L. Cerebral glucose metabolic rates in normal human females versus normal males. *Psychiatry Res*. 1987;21:237-245.
123. Andreason PJ, Zametkin AJ, Guo AC, Baldwin P, Cohen RM. Gender-related differences in regional cerebral glucose metabolism in normal volunteers. *Psychiatry Res*. 1993;51:175-183.
124. Berman KF, Schmidt PJ, Rubinow DR, et al. Modulation of cognition-specific cortical activity by gonadal steroids: a positron-emission tomography study in women. *Proc Natl Acad Sci U S A*. 1997;94:8836-8841.
125. Shaywitz SE, Shaywitz BA, Pugh KR, et al. Effect of estrogen on brain activation patterns in postmenopausal women during working memory tasks. *JAMA*. 1999;281:1197-1202.
126. Wong DF, Broussolle EP, Wand G, et al. In vivo measurement of dopamine receptors in human brain by positron emission tomography: age and sex differences. *Ann N Y Acad Sci*. 1988;515:203-214.
127. Smith MJ, Keel JC, Greenberg BD, et al. Menstrual cycle effects on cortical excitability. *Neurology*. 1999;53:2069-2072.
128. Smith MJ, Adams LF, Schmidt PJ, Rubinow DR, Wassermann EM. Ovarian hormone effects on human cortical excitability. *Ann Neurol*. 2002. In press.
129. von Feuchtersleben E. *The Principles of Medical Psychology*. London, UK: Sydenham Society; 1847.
130. Rubinow DR, Roy-Byrne PP. Premenstrual syndromes: overview from a methodologic perspective. *Am J Psychiatry*. 1984;141:163-172.
131. Rubinow DR, Roy-Byrne PP, Hoban MC, Gold PW, Post RM. Prospective assessment of menstrually related mood disorders. *Am J Psychiatry*. 1984;141:684-686.
132. NIMH Premenstrual Syndrome Workshop Guidelines. Rockville, Md: National Institute of Mental Health; 1983.
133. American Psychiatric Association. *Diagnostic and Statistical Manual of Mental Disorders*. 3rd ed, revised. Washington, DC: American Psychiatric Association; 1987.
134. Wang M, Seippel L, Purdy RH, Bäckström T. Relationship between symptom severity and steroid variation in women with premenstrual syndrome: study on serum pregnenolone, pregnenolone sulfate, 5α-pregnane-3,20-dione and 3α-hydroxy-5α-pregnan-20-one. *J Clin Endocrinol Metab*. 1996;81:1076-1082.
135. Redei E, Freeman EW. Daily plasma estradiol and progesterone levels over the menstrual cycle and their relation to premenstrual symptoms. *Psychoneuroendocrinology*. 1995;20:259-267.
136. Facchinetti F, Genazzani AD, Martignoni E, Fioroni L, Nappi G, Genazzani AR. Neuroendocrine changes in luteal function in patients with premenstrual syndrome. *J Clin Endocrinol Metab*. 1993;76:1123-1127.
137. Backstrom T, Aakvaag A. Plasma prolactin and testosterone during the luteal phase in women with premenstrual tension syndrome. *Psychoneuroendocrinology*. 1981;6:245-251.
138. Eriksson E, Sundblad C, Lisjo P, Modigh K, Andersch B. Serum levels of androgens are higher in women with premenstrual irritability and dysphoria than in controls. *Psychoneuroendocrinology*. 1992;17:195-204.
139. Bloch M, Schmidt PJ, Su TP, Tobin MB, Rubinow DR. Pituitary-adrenal hormones and testosterone across the menstrual cycle in women with premenstrual syndrome and controls. *Biol Psychiatry*. 1998;43:897-903.
140. Schechter D, Strasser TJ, Endicott J, Petkova E, Nee J. Role of ovarian steroids in modulating mood in premenstrual syndrome. *Abstr Soc Biol Psych 51st Annu Meeting*. 1996:646.
141. Halbreich U, Endicott J, Goldstein S, Nee J. Premenstrual changes and changes in gonadal hormones. *Acta Psychiatr Scand*. 1986;74:576-586.
142. Roca CA, Schmidt PJ, Bloch M, Rubinow DR. Implications of endocrine studies of premenstrual syndrome. *Psychiatr Ann*. 1996;26:576-580.
143. Rubinow DR, Schmidt PJ, Roca CA, Daly RC. Gonadal hormones and behavior in women: concentrations vs context. In: Pfaff D, ed. *Hormones, Brain and Behavior*. San Diego, Calif: Academic Press. 2002. In press.
144. Schmidt PJ, Nieman LK, Grover GN, Muller KL, Merriam GR, Rubinow DR. Lack of effect of induced menses on symptoms in women with premenstrual syndrome. *N Engl J Med*. 1991;324:1174-1179.
145. Schmidt PJ, Nieman LK, Danaceau MA, Adams LF, Rubinow DR. Differential behavioral effects of gonadal steroids in women with and in those without premenstrual syndrome. *N Engl J Med*. 1998;338:209-216.
146. Bancroft J, Boyle H, Warner P, Fraser HM. The use of an LHRH agonist, buserelin, in the long-term management of premenstrual syndromes. *Clin Endocrinol*. 1987;27:171-182.
147. Mortola JF, Girton L, Fischer U. Successful treatment of severe premenstrual syndrome by combined use of gonadotropin-releasing hormone agonist and estrogen/progestin. *J Clin Endocrinol Metab*. 1991;71:252A-252F.
148. Rubinow DR, Hoban MC, Grover GN, et al. Changes in plasma hormones across the menstrual cycle in patients with menstrually related mood disorder and in control subjects. *Am J Obstet Gynecol*. 1988;158:5-11.
149. Rubinow DR, Schmidt PJ, Roca CA. Estrogen–serotonin interactions: implications for affective regulation. *Biol Psychiatry*. 1998;44:839-850.
150. Ho HP, Olsson M, Westberg L, Melke J, Eriksson E. The serotonin reuptake inhibitor fluoxetine reduces sex steroid–related aggression in female rats: an animal model of premenstrual irritability? *Neuropsychopharmacology*. 2001;24:502-510.
151. Bancroft J, Cook A, Davidson D, Bennie J, Goodwin G. Blunting of neuroendocrine responses to infusion of L-tryptophan in women with perimenstrual mood change. *Psychol Med*. 1991;21:305-312.
152. Su TP, Schmidt PJ, Danaceau M, Murphy DL, Rubinow DR. Effect of menstrual cycle phase on neuroendocrine and behavioral responses to the serotonin agonist *m*-chlorophenylpiperazine in women with premenstrual syndrome and controls. *J Clin Endocrinol Metab*. 1997;82:1220-1228.
153. Dimmock PW, Wyatt KM, Jones PW, O'Brien PMS. Efficacy of selective serotonin-reuptake inhibitors in premenstrual syndrome: a systematic review. *Lancet*. 2000;356:1131-1136.
154. Menkes DB, Coates DC, Fawcett JP. Acute tryptophan depletion aggravates premenstrual syndrome. *J Affect Disord*. 1994;32:37-44.
155. Roca CA, Schmidt PJ, Smith MJ, Danaceau MA, Murphy DL, Rubinow DR. Effects of metergoline on symptoms in women with premenstrual dysphoric disorder. *Am J Psychiatry*. 2002. In press.
156. Beilin J, Zajac JD. Function of the human androgen receptor varies according to CAG repeat number within the normal range. 81st Annual Meeting Endocrinology Society. San Diego, Calif. 1999;500. Abstract.

157. Kieback DG, Tong XW, Weigel NL, Agoulnik IU. A genetic mutation in the progesterone receptor (PROGINS) leads to an increased risk of non-familial breast and ovarian cancer causing inadequate control of estrogen receptor driven proliferation. *J Soc Gynecol Invest*. 1998;5:40a.
158. Giovannucci E, Stampfer MJ, Krithivas K, et al. The CAG repeat within the androgen receptor gene and its relationship to prostate cancer. *Proc Natl Acad Sci U S A*. 1997;94:3320-3323.
159. Wang-Gohrke S, Chang-Claude J, Becher H, Kieback DG, Runnebaum IB. Progesterone receptor gene polymorphism is associated with decreased risk for breast cancer by age 50. *Cancer Res*. 2000;60:2348-2350.
160. Spearow JL, Doemeny P, Sera R, Leffler R, Barkley M. Genetic variation in susceptibility to endocrine disruption by estrogen in mice. *Science*. 1999;285:1259-1261.
161. Lesch KP, Bengel D, Heils A, et al. Association of anxiety-related traits with a polymorphism in the serotonin transporter gene regulatory region. *Science*. 1996;274:1527-1531.
162. Pollock BG, Ferrell RE, Mulsant BH, et al. Allelic variation in the serotonin transporter promoter affects onset of paroxetine treatment response in late-life depression. *Neuropsychopharmacology*. 2000;23:587-590.
163. Smith SS, Gong QH, Hsu FC, Markowitz RS, ffRench-Mullen JMH, Li X. GABAA receptor alpha-4 subunit suppression prevents withdrawal properties of an endogenous steroid. *Nature*. 1998;392:926-930.
164. Follesa P, Serra M, Cagetti E, et al. Allopregnanolone synthesis in cerebellar granule cells: roles in regulation of $GABA_A$ receptor expression and function during progesterone treatment and withdrawal. *Mol Pharmacol*. 2000;57:1262-1270.
165. Smith SS, Gong QH, Li X, et al. Withdrawal from 3α-OH-5α-pregnan-20-one using a pseudopregnancy model alters the kinetics of hippocampal $GABA_A$-gated current and increases the $GABA_A$ receptor α4 subunit in association with increased anxiety. *J Neurosci.* 1998;18:5275-5284.
166. Ströhle A, Romeo E, Hermann B, et al. Concentrations of 3α-reduced neuroactive steroids and their precursors in plasma of patients with major depression and after clinical recovery. *Biol Psychiatry*. 1999;45:274-277.
167. Romeo E, Brancati A, de Lorenzo A, et al. Marked decrease of plasma neuroactive steroids during alcohol withdrawal. *Clin Neuropharmacol*. 1996;19:366-369.
168. Bitran D, Hilvers RJ, Kellogg CK. Anxiolytic effects of 3α-hydroxy-5α[β]-pregnan-20-one: endogenous metabolites of progesterone that are active at the $GABA_A$ receptor. *Brain Res*. 1991;561:157-161.
169. Purdy RH, Morrow AL, Moore PH, Jr, Paul SM. Stress-induced elevations of gamma-aminobutyric acid type A receptor–active steroids in the rat brain. *Proc Natl Acad Sci U S A*. 1991;88:4553-4557.
170. Griffin LD, Mellon SH. Selective serotonin reuptake inhibitors directly alter activity of neurosteroidogenic enzymes. *Proc Natl Acad Sci U S A*. 1999;96:13512-13517.
171. Schmidt PJ, Purdy RH, Moore PH, Jr, Paul SM, Rubinow DR. Circulating levels of anxiolytic steroids in the luteal phase in women with premenstrual syndrome and in control subjects. *J Clin Endocrinol Metab*. 1994;79:1256-1260.
172. Daly RC, Bloch M, Rubinow DR, Kim HY, Schmidt PJ. Neurosteroids in an endocrine model for postpartum mood disorders. 154th American Psychiatric Association Meeting. New Orleans, La. 2001;191. Abstract.
173. Meltzer ES, Kumar R. Puerperal mental illness, clinical features and classification: a study of 142 mother-and-baby admissions. *Br J Psychiatry*. 1985;147:647-654.
174. Wisner KL, Peindl K, Hanusa BH. Relationship of psychiatric illness to childbearing status: a hospital-based epidemiologic study. J Affect Disord. 1993;28:39-50.
175. Wisner KL, Peindl KS, Hanusa BH. Psychiatric episodes in women with young children. *J Affect Disord*. 1995;34:1-11.
176. Cutrona CE. Causal attributions and perinatal depression. *J Abnorm Psychol*. 1983;92:161-172.
177. Kumar R, Mordecai Robson K. A prospective study of emotional disorders in childbearing women. *Br J Psychiatry* . 1984;144:35-47.
178. Cox JL, Murray D, Chapman G. A controlled study of the onset, duration and prevalence of postnatal depression. *Br J Psychiatry*. 1993;163:27-31.
179. Harris B, Huckle P, Thomas R, Johns S, Fung H. The use of rating scales to identify post-natal depression. *Br J Psychiatry*. 1989;154:813-817.
180. Harris B, Lovett L, Newcombe RG, Read GF, Walker R, Riad-Fahmy D. Maternity blues and major endocrine changes: Cardiff puerperal mood and hormone study II. *BMJ*. 1994;308:949-953.
181. Harris B. Biological and hormonal aspects of postpartum depressed mood: working towards strategies for prophylaxis and treatment. *Br J Psychiatry*. 1994;164:288-292.
182. Cooper PJ, Campbell EA, Day A, Kennerley H, Bond A. Non-psychotic psychiatric disorder after childbirth: a prospective study of prevalence, incidence, course and nature. *Br J Psychiatry*. 1988;152:799-806.
183. Evans J, Heron J, Francomb H, Oke S, Golding J. Cohort study of depressed mood during pregnancy and after childbirth. *BMJ*. 2001;323:257-260.
184. Josefsson A, Berg G, Nordin C, Sydsjo G. Prevalence of depressive symptoms in late pregnancy and postpartum. *Acta Obstet Gynecol Scand*. 2001;80:251-255.
185. Cox JL, Connor Y, Kendell RE. Prospective study of the psychiatric disorders of childbirth. *Br J Psychiatry*. 1982;140:111-117.
186. Kendell RE. Emotional and physical factors in the genesis of puerperal mental disorders. *J Psychosom Res*. 1985;29:3-11.
187. O'Hara MW. Social support, life events, and depression during pregnancy and the puerperium. *Arch Gen Psychiatry*. 1986;43:569-573.
188. Frank E, Kupfer DJ, Jacob M, Blumenthal SJ, Jarrett DB. Pregnancy-related affective episodes among women with recurrent depression. *Am J Psychiatry*. 1987;144:288-293.
189. Whiffen VE. Vulnerability of postpartum depression: a prospective multivariate study. *J Abnorm Psychol.* 1988;97:467-474.
190. Watson JP, Elliott SA, Rugg AJ, Brough DI. Psychiatric disorder in pregnancy and the first postnatal year. *Br J Psychiatry*. 1984;144:453-462.
191. Pitt B. "Atypical" depression following childbirth. *Br J Psychiatry*. 1968;114:1325-1335.
192. Cooper PJ, Murray L. Course and recurrence of postnatal depression evidence for the specificity of the diagnostic concept. *Br J Psychiatry*. 1995;166:191-195.
193. Dean C, Williams RJ, Brockington IF. Is puerperal psychosis the same as bipolar manic-depressive disorder? A family study. *Psychol Med*. 1989;19:637-647.
194. Hobfoll SE, Ritter C, Lavin J, Hulsizer MR, Cameron RP. Depression prevalence and incidence among inner-city pregnant and postpartum women. *J Consult Clin Psychol*. 1995;63:445-453.
195. Hamilton JA, Parry BL, Blumenthal SJ. The menstrual cycle in context, I: affective syndromes associated with reproductive hormonal changes. *J Clin Psychiatry*. 1988;49:474-480.
196. Bloch M, Daly RC, Rubinow DR. Endocrine factors in the etiology of postpartum depression. *Compr Psychiatry*. 2002. In press.
197. Pearlstein TB, Frank E, Rivera-Tovar A, Thoft JS, Jacobs E, Mieczkowski TA. Prevalence of axis I and axis II disorders in women with late luteal phase dysphoric disorder. *J Affect Disord*. 1990;20:129-134.
198. Dennerstein L, Morse C, Gotts G, et al. Perspective from a PMS clinic. In: Gise LH, Kase NG, Berkowitz RL, eds. *The Premenstrual Syndromes*. New York, NY: Churchill Livingstone; 1988:109-118.
199. McGill H, Burrows VL, Holland LA, Langer HJ, Sweet MA. Postnatal depression: a Christchurch study. *N Z Med J*. 1995;108:162-165.
200. Sugawara M, Toda MA, Shima S, Mukai T, Sakakura K, Kitamura T. Premenstrual mood changes and maternal mental health in pregnancy and the postpartum period. *J Clin Psychol*. 1997;53:225-232.
201. Gregoire AJP, Kumar R, Everitt B, Henderson AF, Studd JWW. Transdermal oestrogen for treatment of severe postnatal depression. *Lancet*. 1996;347:930-933.
202. Sichel DA, Cohen LS, Robertson LM, Ruttenberg A, Rosenbaum JF. Prophylactic estrogen in recurrent postpartum affective disorder. *Biol Psychiatry*. 1995;38:814-818.
203. Salo LK, Makela SI, Stancel GM, Santti RS. Neonatal exposure to diethylstilbesterol permanently alters the basal and 17 beta-estradiol induced expression of c-fos proto-oncogene in mouse urethroprostatic complex. *Mol Cell Endocrinol*. 1997;126:133-141.
204. vom Saal FS. Sexual differentiation in litter-bearing mammals: influence of sex of adjacent fetuses in utero. *J Anim Sci*. 1989;67:1824-1840.