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Gender Differences in Brain and Behavior: Hormonal and Neural Bases

SANDRA J. KELLY,* NANCY L. OSTROWSKI,† AND MARLENE A. WILSON‡

Department of Psychology and †Department of Pharmacology & Physiology, School of Medicine, University of South Carolina, Columbia, SC 29208, and ‡Eli Lilly and Co., Women's Health Unit, Indianapolis, IN 46285

KELLY, S. J., N. L. OSTROWSKI AND M. A. WILSON. *Gender differences in brain and behavior: Hormonal and neural bases.* PHARMACOL BIOCHEM BEHAV **64**(4) 655–664, 1999.—This article briefly discusses the difficulties in determining the brain–behavior relationship and reviews the literature on some potential mechanisms underlying gender differences in behavioral responses. Mechanisms that are discussed include genetic effects, organizational effects of gonadal hormones, genomic actions of steroids, nongenomic effects of steroids, and environmental influences. The review is an introduction to the articles presented in this special volume on gender differences in brain and behavior. © 1999 Elsevier Science Inc.

Gender differences Sex differences Estrogen Hormones Review

THE articles in this volume report results of investigations of gender or sex differences in behavior, predominantly in rodents. Here, the terms "gender" and "sex" are used interchangeably. Sexually dimorphic behaviors in mammals can be considered the end result of reciprocal influences among genes, gonadal sex, hormonal sex, organizational and activational effects of hormones on the brain, trophic actions of hormones, learning, and social and other environmental influences. Most often, in mammals, these aspects of sex are aligned. However, there are both naturally occurring anomalies and devised experimental situations where these aspects become dissociated, and these conditions have done much to facilitate our understanding of gender differences.

There is ample evidence of gender differences in basic neural processes and behaviors, yet the majority of research using rodents is done exclusively on males. The value of using both sexes in research is that general brain mechanisms underlying behavior are frequently illuminated by the differences between genders. Gender differences are usually the result of complex interactions between the simple direct, temporary influences of fluctuating gonadal hormones known as activational effects, and the permanent organizational influences of steroid hormones. Many behaviors reflect the interdependence of different types and/or levels of sexual dimorphisms. The purpose of this article is to briefly review the different ways in which gender differences in behavioral responses can arise, and review some of the factors that influence gender differences in behavior. The discussion begins with the different forms that gender differences can take, some of the difficulties in the assessment of gender differences in behavior, and their relationship to sexual dimorphisms in brain. Then the focus shifts to how gender differences arise during development, during adulthood, and from environmental events.

CONSIDERATIONS IN THE ASSESSMENT OF GENDER DIFFERENCES

Experimental methods aimed at investigating sex differences use a variety of approaches including gonadectomy followed by hormone replacement, correlational studies, lesion experiments, pharmacological challenges, and assessment of fluctuations over the reproductive cycle. In general, however, complex responses fail to show sex differences that are clearly eliminated by gonadectomy in adulthood and reinstated by gonadal hormone treatments [e.g., see Rivier, this volume, (123)]. Further, although a gonadal hormone treatment in a gonadectomized animal may dramatically modify any particular response, it cannot be assumed that such an effect will represent itself as a robust, gender-related difference. The effects of exogenous administration of a single hormone might be consistent with the hypothesis that there are gender differences in a behavior, but are not proof that a gender difference in intact animals will be observed. Growing evidence suggests

Requests for reprints should be addressed to Sandra J. Kelly, Department of Psychology, University of South Carolina, Columbia, SC 29208.

that this may, in part, reflect complex interactions between genomic and nongenomic actions of gonadal steroids.

On occasion, it has been possible to link specific structural and functional differences in the brain with sexually dimorphic behaviors. For example, the administration of estrogen to gonadectomized females leads, hours later, to increased dendritic spines and synaptic connectivity in the ventromedial hypothalamic nucleus (VMN) (45,85), and induces the expression of progesterone and oxytocin receptors in specific brain sites (7,24,25,74,118,127). Following progesterone or oxytocin administration, females display lordosis in response to a male (14,112). The male VMN does not respond to the same degree to hormone treatments, and males do not demonstrate lordosis in response to a male following a feminine hormone regimen. Based on these and other experiments, the gender-specific, hormone-induced structural changes in the VMN are presumed to underlie the expression of lordosis by females in response to a male. Thus, in the presence of well-defined end points such as lordosis, and clearly identifiable dimorphic brain regions, it becomes possible to establish brain–behavior relationships.

More often than not, however, the relationship between brain and behavior is less than well defined. For example, human males demonstrate greater brain hemispheric asymmetries, have larger sexually dimorphic areas within the hypothalamus, and a differently shaped corpus callosum than females (2–4,32,55). However, it remains controversial what role the sexually dimorphic areas within the hypothalamus play in determining behavior, and whether sex differences in the corpus callosum have behavioral significance (5,32,63). It may be that the sex differences in brain are shaping the same behavior. For example, both sexes show stress-induced analgesia, but pharmacological studies suggest that distinct neurochemical pathways may mediate this behavior in males and females (95). On the other hand, sometimes sex differences in behavior are difficult or impossible to detect or measure because the sensitivity of the behavioral measure may be insufficient to detect a difference between the genders.

Understanding sex differences in brain–behavior relationships of complex behaviors poses a particularly difficult problem. This is because our measures of complex behaviors are influenced by many factors such as sensory processing, motor speed, attention, response to stress, etc., which are sexually dimorphic or influenced by gonadal hormones. Female rats show increased locomotion, enhanced limb coordination, improved sensory perception, and enhanced attentional mechanisms on the night of behavioral estrus. Such hormone-induced effects can influence observations in complex learning and memory tasks or tests of anxiety. For example, results of tests for sex differences in anxiety-related behaviors assessed using different animal models are inconsistent. This is most likely not due to differences in "anxiety" levels per se, but related to the manner in which a particular test measures an animal's level of anxiety. As pointed out by Steenbergen et al. (136) various tests examine different, sometimes conflicting tendencies, including activity related to exploration of novel environments, avoidance of some aspect of the animal's environment, and general activity levels. Thus, female rodents tend to show reduced levels of anxiety in the open-field test, elevated plus-maze task, and the social interaction test (1,59,62,125,162), while other tests demonstrate higher anxiety-related measures in females, or a sex difference that is dependent upon the stage of the estrous cycle (42). Males may be more greatly affected by prior experience than females [discussed in more detail in Fernandes et al., this issue; (1,41,62,136)]. This point has long been debated, but contributing factors to sex differences in several anxiety tests are differences in overall activity levels, responses to specific testing conditions and shock sensitivity (11). Clearly, before a relationship between brain differences and behavior can be established, a very complete analysis of the complex behavior itself is necessary.

One method of elucidating the underlying sexual dimorphisms in the brain associated with complex behaviors is to present a challenge to an organism and assess the response. Challenges include drug administration, perturbations to homeostasis, physical trauma, or the controlled exposure to stressors. Measurements of receptor sensitivity, responses to drugs, changes in behavioral endpoints, etc., often reveal sex differences that are not apparent using baseline measures. For example, animal studies have demonstrated sex differences in sensitivity to several anxiolytic drugs including benzodiazepines such as diazepam (42), serotonin 5-HT_{1a} agents (22), and ethanol [see Devaud et al., Rivier, this volume (34,123)], although the direction of the sex differences in sensitivity to these compounds is inconsistent. Changes in reproductive status or administration of gonadal hormones also change sensitivity to anxiolytics in humans (37,70,137) and animals (18,42,103), suggesting activational control of sensitivity to at least some of these compounds. Challenge situations present a unique set of difficulties in that both sex differences in the processing of the challenging stimulus, as well as sex differences in the response to the stimulus must be considered (155). For example, as indicated above, exploration of gender differences using pharmacological challenges must consider the ability of gonadal hormones to modify the pharmacokinetics (metabolism and distribution) of the drug [e.g., (104)], the integrity of the subsystems on which the drug acts (e.g., neurotransmitter receptor densities), and the neural output pathways involved in the response. Studies that have achieved similar drug levels [e.g., (28,105)], or have used routes of administration that bypass differences due to drug metabolizing enzymes (9), suggest that gonadal hormones and gender modulate sensitivity to a variety of compounds. Such challenge studies can also suggest sex differences in various neurotransmitter systems that contribute to dimorphisms in related integrated behavioral responses.

Despite these difficulties, considerable progress has been made toward understanding the endocrine and neural bases for various gender differences in behavior. The neural substrates underlying gender differences frequently involve the interaction of hormonal events during development, hormonal action during adulthood, and environmental events. For the purposes of organization, these mechanisms of gender differences will be discussed separately.

GENDER DIFFERENCES AND DEVELOPMENT

One of the fundamental differences between the mammalian sexes is the expression of genes on the Y chromosome, whose protein products promote differentiation of the primordial gonads in the testes in the fetal male (106). This, in turn, gives rise to developmental hormonal events that result in male development. However, there is some evidence that there may be other chromosomal/genetic events that are independent of the testes that also result in male differentiation. For example, in some species, sexual differentiation and determination of sex ratios occurs before gonad determination (128,140). In addition, there are differences in how the maternal and paternal genomes contribute to the develop-

ment of brain regions and these differential contributions may result in differences between the sexes (69). Demonstrations that some sexually dimorphic brain regions, such as the hypothalamus, appear to develop along gender-specific lines without the stimulus of gonadal hormones suggest that there are other sex-specific genetic mechanisms for differentiation (15). However, most of the mechanisms of gender differences that arise during development involve the organizational effects of the gonadal hormones.

To a great extent, hormonal sex determines the sexual dimorphism of the brain. In rats, gonadal hormones influence sexual differentiation of the grain from gestational day 18 through the second week after birth (81). In early studies of hormonal effects on brain and behavior, investigators found that during species-specific critical periods male hormones, such as testosterone, or metabolites of testosterone [dihydrotestosterone (DHT) and estradiol] masculinized the brain, hypothalamic–pituitary–gonadal axis, and behavior, which laid the foundation for species-typical sexual responses as adults. In the absence of masculinizing hormones, the brain developed as female. For example, Barraclough and Gorski (8) demonstrated testosterone propionate delivered to female rats at 5 days of age permanently impaired the cyclic expression of sexual receptivity. Female rats were not only sterile, anovulatory, and in persistent estrus, but did not exhibit sexual responsivity to the male. Female receptivity could not be induced with estrogen and progesterone, suggesting that the neonatal treatments permanently altered the sensitivity of the brain. Similar neonatal hormone exposure produced no effects in males (56). These early effects of hormones are regulated by a combination of factors including the distribution of nuclear receptors and their subtypes (71), the local presence of aromatase or alpha-reductase, the DNA sequence to which the hormone–receptor complexes bind, and by transcriptional cofactors that can modify the actions of the receptor on gene transcription (65). Gonadal hormones masculinize and defeminize the brain, spinal cord, peripheral target organs, and behavior, in part, by regulating both structure and function.

Sex differences in brain structure appear to be, in part, dependent upon the organizational influences of gonadal hormones. In the 1970s, Raisman and Field (119) demonstrated that females had greater numbers of spine synapses in the preoptic area (POA), a region known to be involved in sexual behavior, than male rats. This difference in adult animals could be reversed by androgen administration during early development. Examination of the rat POA by Gorski and associates subsequently revealed a sexually dimorphic nucleus that was approximately five times as large in males as in females, termed the sexually dimorphic nucleus of the preoptic area (SDN-POS) (53). Consistent with the organizational hypothesis of brain differentiation, early castration of male rats reduced the size of this nucleus, while steroid treatment (testosterone or estrogen, but not androgenic metabolites of testosterone) of females during development increased the size of the SDN-POA. Hormone treatment in adulthood had no effect (52). This sex difference is established by events seen on day 18 of gestation and is reflected by increased neurogenesis of SDN-POA neurons (11,61), decreased cell death (31), and increased dendritic growth (54) in males.

Regions not intimately involved in reproductive control may also be influenced by the organizational influences of hormones. The hippocampus serves as a good example. This region is involved with learning and memory tasks, cognition, and stress responsivity, all of which show sexual dimorphisms. Animal data also suggest that females perform spatial tasks involving global spatial cues less well at proestrus when estrogen levels are highest [(89) for a review]. Further, males and females show different learning curves in the Morris water maze, with males outperforming females. Organizational effects are suggested by the ability of castration in neonatal male rats to produce female learning patterns in adults, and estradiol treatment of neonatal females to produce male-typical learning patterns (151). Estrogen has also been shown to induce synapse formation in hippocampal CA1 neurons in rats, which is thought to be determined by the neonatal actions of testosterone during development. Estrogen treatment of adult males fails to produce the same increase in synaptogenesis as in females. If the aromatization of testosterone to estradiol is blocked during development, however, males demonstrate a female-like response to estradiol treatment in adulthood with similar increases in spine synapses (75). More recent data in mice lacking estrogen receptor α (ERα) also suggest that this receptor may mediate the effects of steroid hormones in a spatial learning task (30). Similarly in humans, Nass and Baker (101) demonstrated significantly enhanced spatial ability in women exposed to excessive androgens during development compared to unaffected female relatives. Theoretically, this excessive exposure to prenatal androgens in females may serve to enhance their spatial ability compared to genetically similar females, and make spatial ability comparable to that found in the male population. Other examples of organizing effects on nonreproduction-related behaviors include gender differences in aggression, running wheel and open-field activity, taste preferences for sweetened and salty solutions, production of and reactions to olfactory stimuli including pheromones, reactivity to noxious stimuli, perceptual characteristics, feeding and weight regulation, rough and tumble play, active avoidance, maze learning, cognition, and memory [see (11) and (55) for reviews].

The organizational effects of hormones are thought to be mediated primarily through genomic steroid effects. The binding of hormones to intracellular steroid receptors initiates a cascade of events contributing to sexually dimorphic brain development and organizational effects. Steroid receptors and their associated protein complexes undergo conformational changes upon binding to steroid hormones. The receptor–hormone complex subsequently binds to specific regulatory regions of target gene promoters. Functional mapping of intracellular steroid receptors show that the DNA binding domains are highly conserved, whereas the carboxyl termini contain more variable sequences that determine hormone binding, dimerization, nuclear localization, interactions with heat shock proteins, and transcriptional activity [see (48)]. The steroid target genes, in turn, alter the organization of the brain, ultimately resulting in the sexually dimorphic behavior. Such changes might range from sex differences in anatomical or neurochemical connections to the modified regulation of gene expression and subsequent protein levels that modulate functional neuronal responses [e.g., see (88)]. The effects of steroids during development can also be indirect, such as those mediated by their effects on the developing serotonergic system (152). The question of how nongenomic (rapid-acting or membrane receptor-mediated) effects of gonadal hormones modulate the organization of the developing brain remains to be addressed.

The neurotrophic effects of gonadal hormones appear to play some role in the developmental determination of sex differences. For example, a role for nerve growth factor (NGF) in the sexual differentiation of the brain is suggested by evidence that the administration of NGF antibodies to neonatal rats prevented testosterone-mediated defeminization of sexu-

ally dimorphic reproductive behavior and excitability of VMN-midbrain projections (159). Similarly, anti-NGF attenuated the estrogen-mediated defeminization of lordosis (57). Additional studies have demonstrated that GAP43 (growth-associated protein, neuromodulin) may play a role in the sexually dimorphic patterns of axonal outgrowth and synaptic connectivity in regions of the hypothalamus (79). Sexually dimorphic patterns of GAP-43 mRNA expression were found during early brain development (55,130). In vitro evidence also supports the trophic influences of gonadal hormones (139). Estrogen induces dose-dependent increases in the frequency of neurite outgrowth, spine development, interneuritic connectivity, gap junction formation and size, and functional dye coupling and the selective estrogen receptor modulator (SERM), raloxifene, mimics estrogen and increases neurite outgrowth (102,139). DHT can also induce dose-dependent increases in mean neurite length, branch order, and neuritic field area. Lustig (78) suggests these results represent the differential effects of estrogens and androgens in vivo, with androgens increasing neurite arborization and receptive field size of individual cells, while complementarily estrogen effects facilitate the actual neural communication by inducing spines, synapses, and gap junctions.

GENDER DIFFERENCES IN ADULTHOOD

Genomic Actions of Steroids in Adults

The administration of steroid hormones to adult rodents elicits behavior patterns that are dependent, to a great extent, on the earlier, sex-specific organizing effects that those hormones had on the neonatal brain. Activational effects of hormones during adulthood can depend on mechanisms involving classic intracellular estrogen, androgen, or progestin receptors that subsequently modify gene expression and ultimately behavior (90,131). These genomic actions of steroids might underlie the multitude of sex differences or influences of gonadal steroid hormones on neuropeptide levels, enzyme levels, and receptor subunit proteins that have been observed [e.g., see (36,88,115,132,154)]. Differences in the transcriptional control of various proteins by gonadal steroids might be critical in determining gender-dependent behaviors relying on these transmitters in specific circuits. These genomically mediated changes in mRNA expression require sufficient time (generally hours to days) and appropriate steroid timing to subsequently modulate protein synthesis (90). Sex differences in expression of estrogen, progestin, or androgen receptors in any particular brain site could underlie a gender-dependent behavioral influence of these steroids. Regionally specific sex differences are seen in the levels or regulation of these steroid receptors (77,80,86,91,160) and in colocalization of steroid receptors with other neurochemicals in some neuronal populations [e.g., (58)]. Of course, many of these differences are observed in brain areas important for control of reproductive functions, and their sexually dimorphic nature is highly relevant to their functions in reproductive behaviors and physiological responses [see (36,132)]. Perhaps the best known example is the ability of estrogen to induce progestin receptors in a regionally selective and sexually dimorphic manner [see (24,25,74,118)]. A recent study, however, did not show sex differences in the estrogen receptor immunoreactivity, or the ability of estrogen administration to downregulate estrogen receptor levels, in hippocampus (148). Estrogen and progestin treatments alter a variety of receptor sites and peptide levels in select regions of hippocampus and many of these responses are sex dependent or (at least) dependent upon the circulating levels of estrogens (88,145–147). Thus, the ability of gender or gonadal hormones to modulate levels of neurotransmitters, peptides, enzymes, or receptors might also be independent of the levels of steroid receptors in a particular brain site. In addition, some of the influences of progesterone can be rapidly mimicked by in vitro incubation of tissue with progesterone derivatives, suggesting potential interactions between the genomic and nongenomic actions of steroids [see below and (26,87,154)].

While estrogen, androgen, and progestins produce multiple changes at the level of the cell, it is often difficult to detect related behavioral change. In rodents, the reliable nocturnal hours of estrus during each estrous cycle provide a window through which hormone-induced behavioral changes can be viewed [e.g., (141)]. In women, such a window is considerably less reliable (55). The human menstrual cycle and hormonal fluctuations are highly variable from woman to woman. Linking effects of hormones to a change in the probability of the occurrence of a specific behavior or the character of that behavior requires accurate measures of hormone levels and sensitive measures of behavior. Certain behaviors may only be sexually dimorphic if measured in females during the days after the preovulatory hormone surge. Finally, some sex differences may require a challenge to the system to measure sexually dimorphic responses. Establishing sex differences in humans still remains a statistical and methodological challenge. With the exception of robust differences in sexual behavior, aggression, and rough and tumble play, most differences reflect a modest statistical shift in the sample measures (e.g., mean, mode, variance, etc.). Clearly, for virtually all end points, both males and females exhibit qualitatively similar behaviors, and vary only with respect to the quantitative aspects of those behaviors.

There are examples of behavioral effects that are at least partially mediated by the activational effects of steroids. The most obvious example is sexual receptivity of female rats, which is dependent upon hormonal changes across the estrous cycle, and clearly involves genomic actions of steroids [see (112)], although nongenomic processes may also be involved (47). Another example includes activity changes in rodents that are sensitive to estrogen levels regardless of sex (51). In humans, there is some suggestion that spatial ability improves with lower estrogen levels, and is worse with high estrogen levels during the midluteal phase (113). Because females may only differ from males for a short time period during their reproductive cycle, the lack of a sex difference in a behavior might not negate the importance of steroid actions on a response, but might suggest that it is limited to a defined point within the cycle. Relatively complex and potentially offsetting influences of gonadal hormones might not be apparent in "baseline" responses, but may become more evidence during situations where the system may require plasticity [e.g., learning and memory; see (142)] or during a pathological insult (e.g., epileptogenesis). Thus, the lack of a sex difference in some response might be related not to the absence of gonadal hormone effects, per se, but a complex set of effects of gonadal hormones that fail to significantly modify basal responding in various measures. In conclusion, observing a gender difference in behavior resulting from activational influences of gonadal hormone might require either a more sophisticated set of measures, a more confined time period in the reproductive cycle, and/or providing some challenge to the system. Further, both a complex set of interactions between hormones and the confinement of effects to defined time points in the female cycle might explain the numerous

contradictions in the literature with respect to sex differences in any particular behavior (e.g., see discussion of anxiety above). Finally, fluctuations over the reproductive cycle do not distinguish genomic and nongenomic actions of steroid hormones, and could also be related to fluctuations in non-steroid hormone effects.

Nongenomic Actions of Steroid Hormones

A growing literature indicates that traditional steroid hormones may also exert nongenomic actions through a variety of interactions with membrane receptors or second messenger cascades. The rapid nature of these hormonal effects on ionic conductance and signaling cascades (within seconds), the inability of classic steroid receptor antagonists to block these effects, and the localization of influences to regions devoid of classic steroid receptors suggest that these effects are not mediated through classic intracellular steroid receptor-mediated changes in transcription. The use of large, BSA-conjugated steroids that cannot cross the membrane have also been used to elucidate physiological changes and demonstrate high-affinity binding that is not associated with classic intracellular steroid receptors (120,138). The recent demonstration that estrogen receptors (both alpha and beta) can be expressed on cellular membranes (121), provides exciting new support for the rather heretical concept of membrane steroid receptors that utilize nongenomic mechanisms to modulate cellular functions [which has been around for many years now (114)]. On the other hand, the interaction of steroids with signaling cascades that can ultimately modulate transcription, might ultimately force further refinement of concurrent definitions of genomic vs. nongenomic effects.

The behavioral consequences of these short-latency, nongenomic actions of steroids are not entirely clear. Nonetheless, nongenomic actions of estrogen in nigrostriatal and cerebellar systems are implicated in modulating locomotor activity and coordination. The ability of estrogen pretreatment to enhance the behavioral effects of dopamine antagonists is thought to be mediated by nongenomic effects on dopaminergic neurons [see Becker this volume; (13,27,135, 158)]. Electrophysiological studies have similarly demonstrated that estrogen appears to have rapid actions on responses to other neurotransmitters, in many instances through nongenomic mechanisms. Such effects can range from altered release to modified postsynaptic effects [see (158)]. Estrogen administration can acutely enhance glutamatergic effects in several brain regions (133–135,156–158), serotonergic responses in the hippocampus (12,38), dopamine responses in the nigrostriatal system [see Becker this issue; (13,27)], opioid responses in the hypothalamus (67), and $GABA_B$ responses in hypothalamic neurons (67). These responses appear to be related to influences on ionic conductances, although some of them may be related to influences on G-protein coupling (67) or neurotransmitter release [see (13,158)]. Estrogen can also alter gene transcription through interaction with the cAMP/protein kinase A signaling cascade and rapidly increases the phosphorylated form of the cAMP response element binding protein (CREB) (143,161). Additional nongenomic actions include estrogen interactions with phospholipase C, phosphoinositide turnover, intracellular pH, calcium influx changes, protein kinase C, tyrosine kinase pathways, G-protein, and ionic conductances [see (144,158)].

Steroid derivatives of progesterone, testosterone, and glucocorticoids, referred to as neuroactive steroids or neurosteroids, can also alter behavioral responses through nongenomic actions, and supports their potential role in mediating sex-related differences. The behavioral actions of neuroactive steroids include sedative/anesthetic, anxiety-reducing, and seizure-reducing properties (17,19,21,44,49,50,96,129,150). These actions of neuroactive steroids are reminiscent of actions of GABA-enhancing compounds such as benzodiazepines, alcohol, and barbiturates. In drug discrimination trials, the subjective effects of neuroactive steroids appear similar (i.e., "generalize") to those of benzodiazepines (6,33). The increased levels of these compounds following acute stress suggests some role of this endocrine response in the neural adaptations associated with responses to stressful stimuli (100,117), and perhaps anxiety-reducing compounds [see Devaud this issue, (34)]. These behavioral responses appear to be related to the ability of these compounds to modulate ligand-gated ionic channels for GABA and glutamate through rapid, direct effects that may not involve classical steroid receptor (i.e., genomic) mechanisms. For example, several steroid metabolites, including the progestin metabolite 3α, 5α-tetrahydroprogesterone (3α,5α -THP; 5α-pregnan-3α -ol-20-one or allopregnanolone, and the androgenic steroids androsterone (5α-androstan-3α ol-17-one) and androstanediol (5α-androstan-3α, 17β diol) have consistently been shown to enhance GABA-mediated responses through direct interaction with a distinct site within the $GABA_A$ receptor complex [see (16,39,82–84, 90,111,154)]. Other neurosteroids, such as pregnenolone sulfate and dehydroepiandrosterone sulfate (DHEAS) antagonize GABA responses or modulate glutamate responses (23,60,82,93,111). Moreover, conditions involving stress or high circulating progestin levels (e.g., pregnancy) appear to produce physiologically relevant brain levels of neuroactive steroids, although the brain has the enzymatic capability of synthesizing some of these compounds (10,20,111,124,135).

Neuroactive steroid brain levels and responses show sex differences and/or modulation by the hormonal milieu. Females have greater basal brain levels of some of these compounds than males and differential levels during different reproductive states (20,116), although overall brain levels do not show striking sex differences following some stressors (153). Neuroactive steroid effects, particularly with respect to their anticonvulsant nature, are generally enhanced by feminine gonadal hormone milieu and in females [see Devaud this issue; (34,35,43,44,46)]. In addition, estrogen treatments can increase the subsequent efficacy of neuroactive steroid effects, suggesting yet another level of complexity to the problem of assessing the role of these neuroactive steroids in gender-related differences in behavioral responses (29,72,73,135). Many of the articles in this issue investigate the potential role these neuroactive steroid derivatives serve in mediating gender-related differences in behavioral responses.

GENDER DIFFERENCES AND THE ENVIRONMENT

Sex differences in animals and humans are shaped not only by biological contributions but also by environmental pressures and experiences. For example, in rats, gerbils, and ferrets, the dams provide more anogenital stimulation to male offspring than to female offspring in the first several postnatal weeks (99). Such stimulation is critical for both sexes in the development of urination and fecal elimination. In addition, the anogenital stimulation provided by mothers to male offspring aids in the development of brain mechanisms that control aspects of male sexual function and ultimately affects male sexual behavior (97). Specifically, anogenital stimulation has been shown to prevent cell death in the spinal nu-

cleus of the bulboca vemosus (98). Interestingly, it has also been found that the high levels of maternal licking in either sex results in dampened hypothalamic–pituitary–adrenal responses to stress (76), which is consistent with the sex differences in stress responsivity. Normally, males receive greater amounts of maternal licking, and have reduced stress responses, compared to females. In this volume, Zimmerberg et al. (163) show how the environmental stressor of maternal separation results in greater enhancement in stress sensitivity in females than males.

There are differences in the treatment of human infants based on sex starting very early. Despite progress in gender equality, mothers of newborns have very strong stereotypes of their children based on sex. For example, Reid (122) found that maternal perceptions of newborn males were that they had broad wide hands, looked tall, large, and athletic, and had a serious temperament. The reverse was found for females. Although they had no documentation of temperament, there were no physical differences between the babies at birth. This study is a replication of a study by Rubin (126). Parke and Sawin (110) found that fathers held their daughters close and snug more frequently and for longer periods during play than they did their sons. Mothers held their sons close more than their daughters. In contrast, for visual attending and stimulation behaviors, fathers favored their sons and mothers favored their daughters. Fathers also made more frequent attempts to stimulate their sons' feeding by moving the bottle than for their daughters, and the reverse was true for mothers. These types of findings have been observed in other cultures (both Israeli kibbutzum and Bushmen of Botswana) and in laboratory studies of wild-born rhesus monkeys (107,109). The long-term effects of these differences are unknown, and may be evocative of other differences later in development.

Environmental insults and complexity during development also clearly interact with gender to produce very different effects. Alcohol exposure during development has repeatedly been shown to have differing effects on the fetus depending upon the sex [see (149) for review]. Usually, the sexually dimorphic effects of a teratogen are the result of interactions of the teratogen with some of the organizational effects of steroids. For example, alcohol has been shown to dampen the testosterone surge in rats (92). As a result, most of alcohol's sexually dimorphic effects are on behaviors that are sexually dimorphic in unexposed animals, and often there is a reversal of the gender difference [e.g., (68,94)]. Some of cocaine's effects [see Katovic et al., this volume, (64)] and diazepam's effects [see Kellogg, this volume (66)] during development have also been shown to be sexually dimorphic. Some environmental insults actually mimic the actions of sex steroids and alter development by altering the organizational events [see Palanza et al., this volume, Farabollini, this volume; (40;108)]. The complexity of the environment can also interact with gender to produce differential effects on the brain. The nature of interaction depends on the brain region and the neuroanatomical component being examined; however, the interaction can be powerful enough to actually reverse the gender difference seen in rats reared in nonenriched environments [see (63) for a review].

Environmental events during development have clear influences on gender differences and the brain; studies of environmental events during adulthood and gender are more often done in the realm of social and clinical psychology and do not have as clear a link to brain function (although certainly one exists). The effect of environmental events or experiences will be determined by existing brain processes, including both genomic and nongenomic effects of steroids, and particular brain processes will also give rise to a higher likelihood of particular experiences or environmental events. The intertwining of environment and brain will clearly be very complex and occur across the lifespan.

CONCLUSIONS

In summary, there is a great amount of literature describing gender differences in the brain and behavior, and multiple mechanisms by which gender differences are induced and measured. The articles in this volume continue the exploration of gender differences in the brain and behavior. It is increasingly important that both genders be included in animal and human research, and that gender-based research contribute to the understanding that sex differences in structure, function, and behavior exist independently of judgments regarding their desirability. In other words, findings in one gender are neither better nor worse, or more or less desirable than our current culture determines. We will need to proceed with the view that gender differences in the brain and behavior will serve as a window into brain function rather than an unwanted complication of future research.

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