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Abstract

Prevalence and symptoms of most psychiatric and neurological disorders differ in men and women and there is substantial evidence that their neurobiological basis and treatment also differ by sex . This special issue sought to bring together a series of empirical papers and targeted reviews to highlight the diverse

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impact of sex in neuroscience and neuropsychopharmacology. This special issue gathers empirical papers and targeted reviews to emphasize the diverse impact of sex in neuroscience and neuropsychopharmacology, including 9 review papers and 17 research articles highlighting investigation in different species (zebra fish, mice, rats and humans). Each contribution covers scientific topics that overlap with genetics, endocrinology, cognition, behavioral neuroscience, neurology and pharmacology. Highlighting the extent to which sex differences can impact the brain and behavior is key to moving forward in neuroscience research.

Introduction

Prevalence and symptoms of most psychiatric and neurological disorders differ in men and women and there is substantial evidence that their neurobiological basis and treatment also differ by sex (Kokras & Dalla, 2014; Shansky & Woolley, 2016; Bale & Epperson, 2017; Joel & McCarthy, 2017; Lee, 2018; Arnegard et al., 2020). Sex differences themselves have been the focus of a growing body of research in Neuroscience and Neuropsychopharmacology world-wide and are evident in most research areas studied to date. For example, sex differences are present in neuroendocrine functions, stress response, cognitive processes, as well as in neuropsychiatric, neurodegenerative, neurodevelopmental disorders and their pharmacological treatments (Dalla et al., 2010; Kokras & Dalla, 2017). The regulation of sex differences by peripheral and central sex hormones is very important and behavioral, electrophysiological, molecular, proteomic, transcriptomic, genetic and neuroimaging data from males and females often differ. Therefore, there is an increasing need to understand how reproductive state and its interaction with developmental stage and aging does affect sex-specific outcomes.

This special issue stems from conversations at the 2018 meeting of the Federation of European Neurosciences (FENS) and the realization that a greater focus on sex differences and women's health is needed in European, as well as world-wide, neuroscience research. It follows the 2015 policies of the National Institutes of Health (NIH, USA NOT-OD-15-102), the Canadian Institutes of Health Research (CIHR/IRSC), Natural Sciences and Engineering Research Council of Canada (NSERC/CRSNG) and European Union policy for Horizon 2020 programs (2016) to incorporate both males and female subjects in all funded research projects with the aim of improving our understanding of sex specific effects in health and disease.

This special issue gathers empirical papers and targeted reviews to emphasize the diverse impact of sex in neuroscience and neuropsychopharmacology, including 9 review papers and 17 research articles highlighting investigation in different species (zebra fish, mice, rats and humans). Each contribution covers scientific topics that overlap with genetics, endocrinology, cognition, behavioral neuroscience, neurology and pharmacology. Authors include world-wide investigators from junior and senior researchers to experts from Europe, Asia, South America, North America and Australia. We thank the authors, the reviewers and the Editorial team at the European Journal of Neuroscience Office for their time and dedication on this special issue. Highlighting the extent to which sex differences do impact the brain and behavior is key to open new perspectives in neuroscience research in sex differences and improve its impact in societies.

Sex differences in Neuroscience and Neuropharmacology

While initial interest on sex differences in the brain focused on the adulthood period, the present issue includes emerging research paradigms that now extend across all stages of the life span, from early brain development, and adolescence, to aging across many domains that are often overlapping, such as cognition, emotion and hormonal effects. Here we have briefly summarized key themes and findings.

Neuroendocrinology: One of the most common areas of research for sex differences has been the investigation of steroid hormones and the hypothalamic-pituitary-gonadal (HPG), or -adrenal (HPA) systems, where estradiol and glucocorticoids were the focus of most of this research to date. For decades, the usual suspects for steroid action and sex difference in the central nervous system were limited to the hypothalamus, however, it became clear that steroids affect many brain regions. In this issue, Willett et al investigate how the estrous cycle modulates the physiological properties of caudate-putamen medium spiny neurons (MSN) and show that MSN of the female caudate-putamen are sensitive to the estrous cycle, pointing to the importance of sex differences and neuroendocrine states in neuron physiology (Willett et al., 2019). This work adds up to the amazing literature on other brain regions where neural cells are modulated by sex hormones, including in the hippocampus (Brandt & Rune, 2019; Yagi & Galea, 2019) and in the prefrontal cortex (Frick et al., 2018). In addition, as pointed by Khbouz et al, the effect of steroids does not only require nuclear receptors and classical intra-cellular pathways, but implicates non-classical membrane receptors, such as membrane ERalpha (mERalpha) receptor in perinatal programming of the brain (Khbouz et al., 2019). Using a mouse model with a mutation in the ERalpha palmitoylation site, they investigated the role of membrane ERalpha in sexual behavior and neuronal populations and reported that mERalpha is particularly important in the development of the male brain (Khbouz et al., 2019).

To add another level of complexity, steroids are not only coming from the peripheral gland but can also be produced centrally. The important effects of local estrogens production in the hippocampus are underscored in the review of Brandt and Rune which summarizes the evidence that local synthesis of estradiol, or neuroestrogens, plays an essential role in synaptic connectivity in the hippocampus of females, but not of male, mice (Brandt & Rune, 2019). Importantly, effects of exogenous estradiol administration, as well inhibition of the enzyme aromatase, which converts androgens to estrogens, are presented in relation to neuroplasticity effects. This review amplifies the need to include "Sex as a Biological Variable" in neuroscience (Brandt & Rune, 2019). Interestingly, the importance of locally produced steroids and sex-dependent impact is also observed in other brain regions usually not perceived as sexually differentiated, including the cortex. Seelers and colleagues demonstrate that centrally produced estrogens regulate radial migration and follow distinct mechanisms in males and females in fetal brain tissue of mice (Sellers et al., 2020).

Sex steroids such as neuroestrogens are not the only hormones to be produced centrally, and, the review of Sze and Brunton focuses on the role of neuroactive steroids in the stress response from early life to puberty and into adulthood (Sze & Brunton, 2019). The theme of stress and sex is further emphasized with the review by Ross and Bockstaele who discuss the importance of catecholamines in modulating the stress response (Ross & Van Bockstaele, 2020). In their review, they propose a triple network model that serves specialized functions important for stress regulation and the emergence of neuropsychiatric disorders. Beyond catecholamines, the authors discuss the importance of crosstalk between a number of brain systems in modulating the impact of stress on psychopathology. Of course, when talking about stress, talking about resilience is equally important. The review by Fallon et al summarizes the state of the knowledge on sex differences in resilience over three domains – i.e., control of the stressor, exercise and treatment with low doses of ketamine. Interestingly, unlike males, behavioral control in female rats does not buffer against outcomes of future adverse events (Fallon et al., 2019). These three reviews on “sex and stress” point out to a number of neural circuits and neurochemicals molecules that should be the focus of future research in stress, stress-related disorders and their treatments.

It should be noted here that when considering neuroendocrine factors, one must also consider the impact of circadian rhythms in health and disease. The importance of sex differences and circadian endocrine rhythms is thoroughly reviewed by Nicolaides and Chrousos with a discussion of historical milestones, including central and peripheral components of the circadian clock system and their clinical implications (Nicolaides & Chrousos, 2020).

Altogether, these articles highlight the importance of sex differences in peripheral and central production of various steroids, their inter-dependency, as well as their sex specific actions on various brain regions, via different molecular pathways. Moreover, it is also important to consider organizational versus activational action of steroids (i.e. actions during development or later in puberty and adulthood). For example, Eck et al., show that limited bedding and nesting during the postnatal period impacts maternal pup-directed care giving behaviors in rodents, as well as masculinization of male offspring, with little effects on female offspring (Eck et al., 2019). This adds up to a growing body of literature on the enduring impact, or organizational impact, of early postnatal environmental stress on later life outcomes. In addition to this, Liu et al show that the stress of isolation via single housing has pervasive effects on cognition, depressive-like behavior and neuroplasticity-related signaling in adult male and female mice, albeit in modest sex-dependent ways (Liu et al., 2019). The sexually differentiated effects of stress are also evidenced in Thompson Gray et al., since stress-induced hyperthermia in a genetic mouse model, SNAP25 Δ 3 mice, results in male mice being more vulnerable than female mice to this stressor (Thompson Gray et al., 2020). During aging, sex frailty is present and Herrera et al show that

following growth factor therapies, behavioral locomotor parameters could be a useful tool to identify sex differences in ageing mice (Herrera et al., 2020).

Cognition, learning and memory: Articles from this issue also highlight the impact of sex differences on cognition and how environmental factors affect cognitive capacities. Using a rodent model, Gogos et al., show that adult male, but not female, offspring exposed to maternal immune activation show deficits in spatial working memory (Gogos et al., 2020). Moreover, Espinosa-Garcia et al., show that everyday diet changes may impact cognition differently in males and females (Espinosa-Garcia et al., 2020). In their rodent model they show that male rats with hypercaloric diet have impairments in spatial learning and memory compared to females, but that long-term memory impairment was evident in both sexes (Espinosa-Garcia et al., 2020). Conner et al., further show that human sex differences in episodic memory can be modeled in rodents and involve specific roles for estrogens and androgens (Conner et al., 2020).

Brain injury: The impact of brain injury comes out as an area where more research on sex differences is needed. While it is now accepted that the impact of brain injury and the following regeneration/degeneration processes are sexually differentiated in adults, mostly due to circulating, but also centrally produced, estrogens and progesterone, current evidence suggests that sex differences also occur early on, prior puberty. This is highlighted in the review of Le Dieu-Lugon et al., that discuss difference in the prevalence and clinical outcomes of encephalopathy of prematurity between sexes in rodent models to understand how these biological mechanisms of this early life condition could aid in developing effective treatments and healthier outcomes for children (Le Dieu-Lugon et al., 2019). Brain injury accounts for millions of emergency room visits per year (CDC, 2019) and the study by Saber et al shows that the mechanisms of traumatic brain injury (TBI) may differ in males and females (Saber et al., 2019). In this study, they used a mouse model of TBI and reported sex differences in interleukin-6 levels as well as sleep duration in the days following the TBI (Saber et al., 2019). Next, Acaz-Fonseca et al., used a rodent model to show that at a cortical steroidogenesis the enzyme aromatase is involved in sex differences in stroke pathophysiology (Acaz-Fonseca et al., 2020). They also show that after ischemic stroke there are cortical sex differences in neuroglobin, sex steroids and autophagy that may be important targets for future research into treatments. Our understanding of cerebral vasculature between the sexes is limited, as highlighted by Oliveira et al., showing that CO administration to adult female mice has a greater impact on cortical structure compared to adult males (Oliveira et al., 2020). These findings have implications not only for brain health, but also for basic processes such as learning and memory.

Neuropsychiatric disorders: Sex differences exist in stress-related disorders, affective disorders, dementia and substance abuse (Balta et al., 2019; Kokras et al., 2019). These neuropsychiatric disorders share neurobiological mechanisms and sex-differentiated features, including genetic, neuroanatomical,

neurochemical, molecular, and hormonal effects that influence the prevalence, the symptomatology, the recovery/relapse, and response to treatment. Neuronal circuits that underlie fear, arousal, anxiety, learning/memory, reward and emotion are also sex-differentiated and contribute to sex differences in neuropsychiatric disorders and neuropsychopharmacology.

In this issue, Day and Stevenson review sex differences in learned fear and its inhibition through extinction and discrimination (Day & Stevenson, 2019). This narrative review points out the need to understand the contribution of endocrine and neural sex differences on learned fear inhibition, which has relevance for understanding affective disorders, such as anxiety.

From the time of adolescence, if not before, understanding the neural basis of anxiety has implications on many aspects of health and disease. Wright et al discuss the need for an integrated translational approach in understanding the neuroscience of social vigilance and social anxiety in adolescent males and females in order to develop effective strategies for treatments (Wright et al., 2019). In keeping with the importance of translational research, Dugandzic et al use a rodent model in their article to show that guanylate cyclase C activation in the amygdala, which is enhanced with feeding (2h after a meal) only in females, is important in the regulation of anxiety-like behaviors and may be an important factor in understanding sex difference in the incidence of anxiety disorders in humans (Dugandzic et al., 2019). Moreover, Fontana et al., report that female zebrafish have higher anxiety-like behavior than males, making zebrafish a valuable model for further understanding the neuroscience of sex differences in anxiety and related disorders (Fontana et al., 2019).

Pharmacology: Medications and drugs of abuse also show marked sex differences in neuroscience research. In the present special issue, sex differences in response to cannabinoids are the focus of research by Poulia et al with findings showing that escalating low Δ^9 -THC doses during adolescence exert differential effects in adult male and female rats, especially when considering the serotonergic system or neuroplasticity (Poulia et al., 2019).

Moreover, Bravo et al showed that there are sex differences in long-lasting behavioral responses following opioid withdrawal, with clear difference between male and female rats. Male animals showed enhanced locomotor effects following a low dose of morphine, while the effect in females was not apparent until high doses of morphine were used (Bravo et al., 2020). This phenomenon is probably related to well-known sex differences in the analgesic properties of opioids.

While the majority of research on sex differences is performed using rodent models, an increasing number of reports confirm the evolutionary conserved sex difference in brain function and sensitivity to pathologies. Indeed, Genario et al., review the literature on sex differences in behavior and

neuropharmacology in zebrafish and point to the importance of zebrafish as a model for continued research in neuropsychopharmacology (Genario et al., 2019).

Conclusions and Future Directions

Sex differences matter and are essential to further the fields of neuroscience and neuropsychopharmacology. In this special issue we emphasize that the impact of sex/gender is pervasive through basic and clinical neuroscience fields. This warrants an increase of awareness of the importance of sex differences in neuroscience and beyond, throughout life and reproductive states to improve our understanding of brain function, neuropsychiatric disorders and treatment efficacy.

As the focus on sex differences in neuroscience fields increases, we hope to see special issues on the impact of sex in specific domains of neuroscience research. Ultimately advocating for research on sex/gender differences will not be necessary, when studying both sexes becomes the norm.

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