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ABSTRACT

Selective serotonin reuptake inhibitor medications (SSRIs) are prescribed to up to 10% of pregnant women to treat maternal mood disorders. Exposure to these medications in-utero has raised concerns about altered neurobehavioral outcomes; most recently those related to peer-to-peer social interactions and play. While clinical data show that both perinatal SSRIs (pSSRI) and maternal stress can contribute to social behavioral changes in children, minimal animal work has investigated the effects of pSSRIs in relevant models of maternal stress or the long-term implications of these effects. Therefore the aim of this work was to investigate the long-term effects of pSSRI exposure to fluoxetine on social behaviors, the hypothalamic pituitary adrenal system (HPA) and hippocampal plasticity in adult male and female rat offspring using a model of pre-gestational maternal stress. Adult Sprague-Dawley female and male rat offspring from the following four groups were utilized: 1.Control + Vehicle, 2. Control + Fluoxetine, 3.Pre-gestational Stress + Vehicle, 4. Pre-gestational Stress + Fluoxetine (n=8-16/female/age groups, n=8-14/male/age groups). Main findings show pSSRIs increased social investigation in adult females and increased social play (pouncing, nape attacks) in adult males. Perinatal SSRIs also had sexually differentiated effects on hippocampal neurogenesis and GR density. Pre-gestational stress had enduring effects by decreasing social investigation and hippocampal neurogenesis in adult males. Thus pSSRIs, as well as pre-gestational maternal stress, have significant long-term effects on social neurobehavioral outcomes which differ in males and females. This suggests that it would be valuable to consider fetal-sex specific treatments for maternal mental illness.

Keywords: Perinatal Depression; SSRI; social behavior; neurogenesis; sex differences; HPA; play

1. Introduction

Selective Serotonin Reuptake Inhibitor medications (SSRIs) are commonly prescribed during pregnancy to treat maternal stress-related mood disorders (Gemmel et al., 2018a; Oberlander et al., 2006). However, perinatal exposure to these medications (pSSRIs) has been linked to poor social development in children highlighted by increased risk of Autism Spectrum Disorder (ASD) (Man et al., 2015) and attention-deficit hyperactivity disorder (ADHD) (Clements et al., 2015) diagnoses. Children prenatally exposed to SSRIs also exhibit low adaptive and social-emotional behavioral scores, and increased externalizing behaviors such as aggression, hyperactivity, and oppositional/defiant behavior (Gemmel et al., 2018a). However, parental depression and poor maternal health in the year prior to conception play an equally, or even more important, role in childhood behavioral outcomes (Brown et al., 2017; Clements et al., 2015; Mezzacappa et al., 2017). Thus, there is a complex relationship between early life exposure to maternal mood disorders and pSSRIs on the development of social behaviors which requires further investigation.

An increasing body of animal research has aimed to understand the effects of pSSRIs on social behaviors. Early-life SSRI treatment reduces social play behaviors including boxing, wrestling, and following/chasing behaviors in juvenile rats (Khatri et al., 2014; Rodriguez-Porcel et al., 2011) as well as social preference via reduced conspecific/object contacts in both males and females in the juvenile and adult period (Khatri et al., 2014; Rodriguez-Porcel et al., 2011; Simpson et al., 2011). However, previous work focused on paradigms in which offspring were directly treated with SSRIs during the postnatal period and, therefore, neglected to consider perinatal exposure-induced outcomes, or outcomes in the context of relevant models of perinatal affective disorders. With this in mind we have shown that in juvenile offspring, pSSRIs prevent the effects of maternal stress on sibling play, but increase aggressive type social play interactions with a novel same-sex stimulus conspecific (Gemmel et al., 2017). Whether these effects persist into adulthood are not known. Interestingly postnatal exposure to SSRIs can increase proceptive behaviors and sexual receptivity in adult female rat offspring while decreasing male

copulatory behaviors (Rayen et al., 2013, 2014), suggesting a long-term impact of pSSRIs on social behaviors in general.

Although limited, there is work documenting sexually-differentiated effects of pSSRIs. Apart from the above mentioned effects on copulatory behaviors, early postnatal SSRI treatment reduces juvenile social preference in male, but not female, rat offspring (Simpson et al., 2011) and increases male stereotypic behaviors, such as self-grooming, during social interaction (Rodriguez-Porcel et al., 2011). Developmental SSRIs also increase immobility in the forced swim in adult female rat offspring (Boulle et al., 2016a, b), regardless of exposure to maternal stress. However, in the absence of a maternal stress paradigm, perinatal SSRI exposure can also increase anxiety and depressive-like behaviors in adult male rodent offspring (Ansorge et al., 2004; Oberlander et al., 2009). It remains to be determined how pSSRIs, in the presence of maternal stress, affect peer-to-peer social interactions in adult *male* and *female* offspring.

Perinatal SSRIs also alter the developing hypothalamic-pituitary-adrenal axis (HPA) (Avitsur, 2017; Avitsur et al., 2016; Pawluski et al., 2012b), a system that has been linked to social interactions and aggressive phenotypes (Walker et al., 2016). For example, unusually low or high HPA axis reactivity is found with the emergence of aggressive behaviors. Work in animal models has shown that pSSRIs have a long-term impact on the HPA system by decreasing corticosterone levels, increasing corticosteroid binding globulin (CBG) levels, altering glucocorticoid receptor density in the hippocampus of offspring, and normalizing the effects of maternal stress on density of dendritic spines and synapses in the hippocampal CA3 region (Gemmel et al., 2017; Ishiwata et al., 2005; Pawluski et al., 2012b). More recently, research shows that pSSRIs, in the absence of maternal stress, alter the ability to respond to continuous stress in both male and female mice offspring (Avitsur, 2017; Avitsur et al., 2016). How these changes in the HPA axis are related to pSSRI effects on social behaviors remains to be determined.

Research has also highlighted the role of the hippocampus in HPA function as well as the formation of social memories pointing to an important role of the CA2 region in this regard (Hammels et

al., 2015; Hitti and Siegelbaum, 2014; Stevenson and Caldwell, 2014). Thus, changes in social behaviors as a result of pSSRIs are likely related to neurobiological changes within this critical limbic region. In pre-adolescent offspring, there was a significant correlation between social interaction with a novel conspecific and the density of glucocorticoid receptors (GR), as well as, pre- and post- synaptic proteins in the CA2 region with additional effects of pSSRIs and pre-gestational maternal stress on neurogenesis and plasticity (Gemmel et al., 2017). Interestingly, the relationship between social interactions and plasticity measures in the CA2 are more pronounced in pre-adolescent female offspring (Gemmel et al., 2017) but it remains to be determined whether these effects endure into adulthood.

The present study aimed to determine the persistent effect of perinatal SSRI exposure to fluoxetine, a popular SSRI prescribed to pregnant women, on social interactions and related changes in the HPA axis and hippocampal plasticity in adult male and female offspring. To determine how changes in social behaviors with a novel same-sex conspecific were linked to the HPA axis, serum levels of corticosterone after social interaction, as well as serum levels of CBG and GR density in the hippocampus and periventricular nucleus (PVN), were assessed. Measures of hippocampal plasticity were also investigated by measuring synaptic proteins in the CA2, CA3, and dentate gyrus and markers of adult neurogenesis in the granule cell layer. A pre-gestational maternal stress paradigm was used as a history of depressive-episodes, anxiety, or prolonged stress prior to pregnancy increase risk of perinatal depression and anxiety (Stewart, 2011). Previous work in animal models shows that pre-gestational chronic unpredictable stress for three weeks immediately prior to gestation can affect both the mother and offspring. For example, pre-gestational stress can affect the developing 5HT system as well as HPA axis reactivity in rat offspring (Gemmel et al., 2017; Gemmel et al., 2018c; Huang et al., 2013; Huang et al., 2010; Huang et al., 2012). Expanding on previous work we expected that pSSRIs would increase social interactions in female offspring, while pre-gestational maternal stress would decrease social behaviors in male offspring, with the exception of social play (pouncing/nape attacks) which has been linked to serotonin levels (Kepser and Homberg, 2015; Kiryanova and Dyck, 2014). We predicted that pSSRI-induced effects on social interactions would be related to alterations in the HPA system as well as the

CA2 region of the hippocampus. Understanding the long-term impact of pSSRIs, in a model of maternal stress/depression, on social interactions and related neuroendocrinology in both male and female offspring is needed to understand the benefits and risks of exposure to antidepressant medications during the perinatal period.

2. Materials and methods

2.1. Animals. Prior to breeding, thirty-four adult female (175-199 g, approximately 60 days of age) and nine adult male (275-299 g) Sprague-Dawley rats from Harlan Laboratories Inc. (Indianapolis, Indiana) were kept in a 12:12-h light/dark schedule under standard laboratory conditions. All experiments were approved by the Institutional Animal Care and Use Committee of Ohio University (IACUC, 12-H-053, 14-H-011) in accordance with the National Institutes of Health guide for the care and use of Laboratory animals (NIH Publications No. 8023, revised 1978).

Three weeks prior to breeding, females were assigned randomly to stress or control groups (16 control, 18 stress). Stressed females were individually housed and subjected to three weeks of chronic unpredictable stress (CUS) consisting of 0-2 stressors per day (restraint stress under bright light, cage rotation, overcrowding, food depravation, wet bedding, forced swim) as previously described (Gemmel et al., 2017) to model aspects of depression and anxiety. For a detailed table of stressors used in the present study see Table 1. Following the 3-week CUS cessation, one female and one male were housed together and gestation day (GD) 1 was identified by a vaginal smear and evidence of sperm in the vagina. Following GD1, all dams were housed individually and monitored daily. The final number of dams that remained pregnant until term were 24 (CV=6, CF=8, PGSV=5, PGSF=5). We have recently shown that this stress paradigm resulted in decreased maternal weight gain, poor maintenance of pregnancy, and altered corticosterone levels in these rat dams (Gemmel et al., 2018b).

2.2. Fluoxetine administration to dams. Fluoxetine (Fagron, Belgium) or vehicle was administered orally via a wafer biscuit from GD10 until weaning (postnatal day (PD) 21). The dams received 1/8th of the

wafer biscuit twice a day filled with vehicle (saline), or 5mg/kg of fluoxetine in vehicle, as described (Gemmel et al., 2017) for a total dose of 10mg/kg/day of fluoxetine. To ensure the dam, and not offspring, consumed the entire biscuit, wafer feeding was completed under supervision.

2.3. Maternal Caregiving Behaviors. To assess any overall effects of maternal care on behavioral outcomes in offspring, maternal caregiving behaviors of offspring were observed in the first week postpartum (PD 1-6), twice per day for 5 min. (Gemmel et al., 2017). Duration of the following maternal behaviors were assessed, and data aggregated across days: licking (licking/grooming), and nursing (arched-back nursing, blanket nursing or passive nursing) to control for any gross effects of maternal care-giving on offspring outcomes. Specific maternal care-giving details in these dams has been recently published and results show that pre-gestationally stressed dams showed higher levels of nursing and fewer bouts of licking/grooming offspring in the first week postpartum (Gemmel et al., 2018b).

2.4. Adult offspring. On PD 1, litters were culled to 4 males and 4 females. Offspring were housed with their mothers until weaning (PD21), then housed with same-sex littermates (4 per cage) and housed 2 per cage at 30 days of age. During adulthood (7-8th postnatal week), a maximum of 2 male and 2 female offspring from a total of 24 litters were used to minimize litter-effects. Offspring from the following treatment groups were used: 1) Control+Vehicle (CV; 10 females, 12 males), 2) Control+Fluoxetine (CF; 14 females, 16 males), 3) Pre-gestational Stress+Vehicle (PGSV; 8 females, 8 males), and 4) Pre-gestational Stress+Fluoxetine (PGSF; 9 females and 10 males).

2.5. Social Interaction Test (SI). Social interaction testing to assess social investigation and play behaviors was completed 24 h following a 15 min habituation to the apparatus during the lights-on portion of the light cycle (between 0900 and 1300). Each experimental animal was paired with a sex- and weight-matched non-experimental stimulus animal bred at our facility specifically to serve as a stimulus partner. A total of 6 male and 6 female stimulus rats were used from 3 litters. These animals were

habituated to the chamber for 10 min prior to testing and were not naïve to social interaction prior to testing with experimental animals.

Sessions were recorded by an overhead digital camera, and videos were evaluated by a blinded observer based on previous work (Crawley, 2012; van Kerkhof et al., 2013; Veenema, 2012). For each experimental animal the duration (s) of the following behaviors were scored: sniffing the stimulus rat; following the stimulus rat; crawling over or under the stimulus rat; grooming the stimulus rat; pouncing on the stimulus rat (experimental rat soliciting the stimulus rat); pinning (the experimental rat holds the stimulus rat on its back); nape attack (the experimental animal displaying nose attacks or nose contacts toward the nape of the neck of the stimulus animal); and running away from the stimulus rat; Self-grooming in the experimental rat was also assessed. Following and sniffing were combined to a single 'social investigation' variable based on previous research (Crawley, 2012; Veenema et al., 2012). In addition, social play behaviors such as pinning, pouncing, and nape-attacks were combined to a single 'social play' variable based on previous work (Veenema et al., 2012). Time until first interaction, total time in each scored behavior, and time interacting were recorded for the experimental animal and can be found in Table 3.

2.6. Corticosterone response to SI. Tail nicks were performed immediately following social interaction testing to obtain blood samples for assessment of corticosterone response to social interaction with a novel conspecific.

2.7. Corticosterone binding globulin (CBG) capacity. At euthanasia (within 24 h of behavioral tests – see below), blood samples were collected and centrifuged at 10,000×g for 10 min, serum separated, and stored at −80 °C. To assess the binding capacity of CBG, samples were diluted 1/1000 for males and 1:1500 for females and incubated with dextran-coated charcoal (DCC) to remove endogenous steroids for 30 min at room temperature. Centrifugation was used to sediment DCC, and 100 µl aliquots were dispensed into duplicate tubes containing [1,2-^{3}H] corticosterone (specific activity: 50 Ci/mmol; ARC, St.

Louis, MO, USA, final concentration ~10 nM) and another tube containing 1 mM cold corticosterone to evaluate non-specific binding. Samples were incubated at room temperature for 1 h, followed by an ice water bath for 30 min. Ice-cold DCC was added for 10 min, and tubes centrifuged at 1800g for 10 min at 4 ºC to remove unbound steroids. To account for the dissociation of [^{3}H] corticosterone from CBG during the latter DCC separation step, a correction factor was used as described (Hammond and Lahteenmaki, 1983). Supernatants were transferred into scintillation vials with 4 ml of Aqueous Counting Scintillant (PerkinElmer, Waltham, MA, USA) to determine the CBG-bound [^{3}H] corticosterone after subtracting non-specifically bound [^{3}H] corticosterone and using this to determine the corticosterone binding capacity of CBG in nM. Inter-assay variability was 5.1%. Estimates of free corticosterone were computed with the equation total corticosterone (nM)*100/CBG(nM) (FCI – free corticosterone index) as previously described (Pawluski et al., 2012a).

2.8. Histology. Within 24 h of SI testing, adult offspring were euthanized (between 1100 and 1500) by deep anesthetization with isofluorane and decapitation. Brains were rapidly dissected, immersion fixed in 4% paraformaldehyde for 48h, and then placed in saturated 30% sucrose solution for approximately a week. Brains were stored at -80°C until coronally sliced by cryostat (Leica Biosystems, Wetzlar, Germany) in 40 micron sections in series of 12. Tissue sections were transferred to a glycol based antifreeze solution and stored at -20°C.

For immunohistochemistry, dorsal hippocampal sections (bregma -2.64 to -4.92) from a random sample of 6 animals per sex per group was used to assess synaptic proteins (PSD-95, synaptophysin) and the glucocorticoid receptor (GR) expression in the CA2, CA3, and dentate gyrus (DG) regions, as well as immature neurons (doublecortin/DCX) in the granule cell layer of the dorsal DG as described (Gemmel et al., 2017). Sections were rinsed between steps in PBS and PBS plus 0.01% Triton X-100 (PBST) for GR staining or TBS and TBS plus 0.01% Triton X-100 (TBST) for all other staining. Tissue was incubated for 30 min at room temperature in 0.6% H_2O_2 followed by blocking for 30 min at room temperature in 5% Normal Goat Serum in PBST or TBST (for GR, synaptophysin, and PSD-95 staining; Lampire Biological

Laboratories, Pipersville PA, USA). For PSD-95 staining, an additional antigen unmasking step was completed for 20 min at 80°C using 10mM sodium citrate buffer (pH 6.0) prior to incubation with H_2O_2. Tissue was then incubated overnight at 4°C in primary goat anti-doublecortin (1:200, Santa Cruz Biotechnology, Dallas Texas, USA), mouse anti-synaptophysin (1:500, Sigma Aldrich, St. Louis MO, USA: NGS+TBST) or rabbit anti-PSD-95 antibody (1:1000, Abcam, Cambridge, MA, USA: NGS+TBST), or in rabbit anti-GR antibody (1:500, Abcam, Cambridge, MA, USA) for two nights. Sections were incubated in appropriate secondary antibody (Vector Laboratories, Burlingame, CA, USA) at room temperature for 2 h followed by processing using the avidin-biotin complex (ABC Elite kit; 1:500; Vector laboratories, Burlingame, CA, USA) and DAB (3,3-diaminobenzidine; Vector laboratories, Burlingame, CA, USA). Sections were mounted on Superfrost Plus slides (Fischer Scientific, Pittsburgh, PA), dried, dehydrated, and cover-slipped with Permount (Fischer Scientific, Pittsburgh, PA). For a representative photomicrograph see Figure 1.

2.9. Quantification. The number of DCX immunoreactive (-ir) cells were counted bilaterally throughout the dorsal GCL/SGZ in 1 series of brain sections (on every 12^{th} hippocampal section) under a 40x objective, as previously described (Barker and Galea, 2008; Gemmel et al., 2017; Gemmel et al., 2015; Gemmel et al., 2016b; Rayen et al., 2015). Cavalieri's principle was used to estimate the total number of DCX-ir cells in the dorsal hippocampus. For optical densities of GR, synaptophysin, and PSD-95-ir cells, two photomicrographs in were taken of each CA2, CA3, and DG/GCL/SGZ region in two sections of the dorsal hippocampus at similar locations between stereotaxic coordinates -2.64 mm to -4.92 mm bregma based on previous work (Gemmel et al., 2017; Gemmel et al., 2015; Pawluski et al., 2012a; Rayen et al., 2015). Photomicrographs for all sections were taken at 40x objective using a Nikon Microphot SA and Nikon DS-Qi1MC camera with Nikon NIS Elements F4.00 software and quantified using Image J software (Wayne Rasband, NIH, Bethesda MD, USA). Two photomicrographs of GR-immunoreactive cells were also quantified for optical density in the paraventricular nucleus (PVN) of the hypothalamus (bregma -1.72 to -1.92). For PVN analysis, photomicrographs were examined under a 40x objective using

(Olympus Provis, equipped with a DP71 digital camera, program Cell ^p) and quantified using Image J64 software as stated above. Quantification was completed by a researcher blind to the conditions.

2.10. Steroid hormones analysis. Blood samples were centrifuged at 10,000×g for 10 min, serum collected, and stored at −80 °C until analysis. For corticosterone levels after SI, samples were run in duplicate using a RIA kit for rat corticosterone from MP Biomedicals (Corticosterone I125 for rats and mice, MP Biomedicals, Santa Ana, CA, USA). The assay had a sensitivity of 7.7 ng and the average coefficient of variation was less than 5%. Corticosterone levels were converted to nM for consistency with the CBG results below by using the equation ng/ml*3.46.

Quantitative determination of serum estradiol in adult female offspring and serum testosterone in adult male offspring (at euthanasia) were completed by enzyme-linked immunosorbent assay following predetermined kit instructions (ALPCO Estradiol Rat ELISA 55-ESTRT-E01, or ALPCO Testosterone Rat/Mouse ELISA 55-TESMS-E01, ALPCO Diagnostics, Salem, NH, USA), with all samples being run in duplicate. Sensitivity of the ELISA was f 2.5 pg/mL for estradiol and 3.06 ng/mL for testosterone. The average coefficient of variation was less than 7%.

2.11. Statistical Analysis. Data were analyzed using the software Statistica (Dell Inc.). Due to previous findings of sexually differentiated patterns of outcomes in the response to pSSRIs and maternal stress exposure on behavioral measures, HPA axis, hippocampal plasticity, and additional neurobehavioral outcomes (Gemmel et al., 2018a; Gobinath et al., 2016; Rayen et al., 2015), factorial ANOVAs were conducted separately for each sex on behavioral measures, corticosterone, CBG, as well as on measures of hippocampal plasticity and GR density with condition (pre-gestational stress/control) and treatment (fluoxetine/vehicle) as independent factors. Factorial ANOVAs with condition (pre-gestational stress/control), treatment (fluoxetine/vehicle), and sex (male/female) as independent factors can be found in Table 2. Significant interaction effects were analyzed by a Fisher LSD *post hoc* test to compare individual group differences. Pearson correlations were analyzed between maternal caregiving behaviors,

corticosterone and CBG measures, social behaviors and neural measurements. Any effects of maternal care-giving behavior or gonadal hormones (estradiol in females and testosterone in males) were also controlled for by using analysis of covariance (ANCOVA) tests. Significance was set at $p<0.05$.

3. Results

3.1. Social Interaction Test. Analysis by sex of social behaviors revealed different patterns of effects in females and males. In adult female offspring, there was a significant main effect of perinatal fluoxetine on time in novel partner-directed social behaviors ($F(1,37)=5.94$, $p=0.02$, Figure 2A) with perinatal fluoxetine exposed females spending more time interacting with a stimulus female. When looking at specific behaviors, perinatal fluoxetine exposed females spent significantly more time engaged in social investigation (main effect of treatment; $F(1,37)=12.34$, $p=0.002$, Figure 2B). There was also a significant effect of perinatal fluoxetine exposure on social grooming (main effect of treatment; $F(1,37)=4.38$, $p=0.04$, Table 3). A significant condition by treatment effect was found with time to first interaction ($F(1,37=6.19$, $p=0.02$, Table 3) with PGSV females taking significantly longer to be socially active compared to CV ($p=0.04$) and PGSF ($p=0.001$) females, while CV females took longer to be socially active compared to PGSF females ($p=0.04$). There was also a significant main effect of treatment on time to first interaction ($F(1,37)=8.74$, $p=0.005$), with fluoxetine exposure reducing time to first interaction in females. There were no significant differences in social play in adult females (Figure 2C) and no other significant differences in social behaviors of adult female offspring.

In adult male offspring, there was a significant main effect of condition on time in novel partner-directed behaviors ($F(1,42)= 6.05$, $p=0.02$, Figure 2D) with males exposed to pre-gestational maternal stress spending less time interacting with a novel stimulus partner. When looking at specific social behaviors, pre-gestationally stressed males spent significantly less time in social investigation with a novel partner compared to control males (main effect of condition; $F(1,42)=18.24$, $p=0.0001$, Figure 2E). In adult male offspring there was also a significant main effect of perinatal fluoxetine treatment on time in

social play (F(1,42)=4.49, p=0.04, Figure 2F) with males exposed to perinatal fluoxetine spending significantly more time in social play than vehicle exposed offspring. Perinatal fluoxetine exposure significantly affected running away from a novel partner (main effect of treatment: F(1,42)=10.63, p=0.002, Table 3) and self-grooming (F(1,42)=5.37, p=0.03, Table 3), with fluoxetine exposed males running away more often and self-grooming more often than vehicle exposed males.

There were no other significant main effects, interaction effects, (Table 3) or correlations and no effect of maternal care-giving or gonadal hormone levels on behavioral outcomes in females or males (ANCOVA, $0.06<p<0.9$).

3.2. HPA Outcomes. In the hippocampus of adult females, there was a significant main effect of treatment (F(1,20)=5.94, p=0.02, Figure 3A) on glucocorticoid receptor density in dentate gyrus, with females exposed to fluoxetine having significantly greater GR density compared to vehicle exposed females. There was also a main effect of condition (F(1,20)=5.01, p=0.04, Figure 3A) on GR density in the dentate gyrus, with adult females exposed to pre-gestational maternal stress having significantly lower density in this region compared to control females. Controlling for adult female estradiol levels diminished the effect of pre-gestational maternal stress on GR density in the DG (ANCOVA; effect of condition; F(1,20)=2.36, p=0.1). There were no other significant effects of condition or treatment on GR density in the hippocampus (Figure 3B, Table 4) or significant correlations between GR density and social behaviors of female offspring (Figure 3C).

In the hippocampus of adult male offspring, there were no significant effects on GR density in the DG, as evident in the female offspring (Figure 3D), but there was a significant main effect of treatment (F(1,20)=4.38, p=0.049, Figure 3E) on GR density in the CA3 region, with males perinatally exposed to fluoxetine having significantly reduced GR density compared to vehicle exposed males. There was also a significant positive correlation between GR density in the DG of adult males and total time in interacting with a novel conspecific (r=0.54, p=0.01, Figure 3F). There were no other significant correlations between GR density, social behaviors or maternal care-giving behaviors in male or female offspring.

In adult females there was a significant main effect of pre-gestational maternal stress on serum corticosterone levels after social interaction ($F(1,34)=4.24$, $p=0.04$, Table 4) and on CBG ($F(1,37)=5.58$, $p=0.02$, Table 4) with adult females exposed to pre-gestational maternal stress having significantly lower corticosterone levels and lower CBG compared to control females. Controlling for estradiol levels diminished the effect of pre-gestational maternal stress on serum corticosterone (ANCOVA; effect of condition; $F(1,34)=2.87$, $p=0.09$).

There were no additional significant effects on free corticosterone, corticosterone or CBG measures in females or males (Table 4) ($p>0.09$). There was no effect of maternal care-giving on HPA measures in females or males (ANCOVA, $0.06<p<1.0$).

3.3. Hippocampal Plasticity. In adult females, there was a significant main effect of treatment on total number of immature neurons in the hippocampal GCL/SGZ ($F(1,20)=4.60$, $p=0.04$, Figure 4A) with perinatal fluoxetine exposure significantly increasing doublecortin-ir. There was also a significant main effect of treatment on post-synaptic density via PSD-95-ir in the CA2 region in adult female offspring ($F(1,20)=5.31$, $p=0.03$, Figure 4B) with perinatal fluoxetine exposure significantly reducing post-synaptic density. There was a non-significant effect of treatment on post-synaptic density in the dentate gyrus ($p=0.06$, Table 5). ANCOVAs revealed no significant effects of estradiol levels on any of the measures ($p>0.6$).

In adult male offspring, there was a significant main effect of condition on total number of immature neurons in the hippocampal GCL/SGZ ($F(1,20)=8.32$, $p=0.009$, Figure 4C) with male offspring exposed to pre-gestational maternal stress having significantly less doublecortin-ir. There was also a significant main effect of treatment on post-synaptic density via PSD-95-ir in the CA2 in adult male offspring ($F(1,20)=6.79$, $p=0.02$, Figure 4D) with perinatal fluoxetine exposure significantly reducing post-synaptic density. There was a significant positive correlation between testosterone levels and synaptophysin density in the dentate gyrus of adult males ($r=0.44$, $p=0.04$, Supplementary Figure 1). There were no other significant differences in adult males or females. There was no effect of maternal care-giving on neural measures in females or males (ANCOVA, $0.09<p<0.96$).

4. Discussion

The current work highlights persistent effects of perinatal fluoxetine exposure, as well as pre-gestational stress, on social behaviors, the HPA axis, and hippocampus plasticity that are sexually differentiated in adulthood. Main findings show that perinatal fluoxetine increased social investigation in adult female offspring and increased social play in adult male offspring. Different patterns of outcomes in male and female offspring, resulting from perinatal fluoxetine exposure, persisted in the hippocampus with regards to GR density and neurogenesis.

4.1. Perinatal SSRIs, Pre-gestational Stress, and Social Behaviors

In the present study perinatal fluoxetine increased social investigation in adult female offspring. Overall, adult females perinatally exposed to fluoxetine were quicker to interact with a novel same-sex conspecific and spent more time engaged in interactions with a novel conspecific. Previous work has shown that perinatal fluoxetine exposure increases female conspecific interaction from postnatal day 28-31 (Svirsky et al., 2016) and facilitates female rat sexual behavior by a significant increase in proceptive behaviors, the lordosis quotient, and decrease in time spent rejecting male advances (Rayen et al., 2014). Interestingly, in pre-adolescent females we found that perinatal fluoxetine did not increase social investigation (Gemmel et al., 2017). This transition to 'pro-social' behaviors in adult female offspring is likely dependent on both organizational effects of pSSRIs and activational effects of gonadal hormones after puberty (Dohler et al., 1991). These 'pro-social' behaviors may be beneficial by increasing social bonds, improving social networks, and ensuring reproductive success. However, it should be noted that clinical work, and animal models, show that pSSRIs are also associated with increased anxiety and depressive-like phenotypes in both males and females (Boulle et al., 2016a; Malm et al., 2016; Olivier et al., 2013).

In addition to effects in adult females, our results show that perinatal fluoxetine exposure increased social play (pinning, nape/attacks), running away, and self-grooming in adult male offspring

when paired with a novel same-sex stimulus conspecific. This expands our previous work in pre-adolescent male offspring which shows that perinatal fluoxetine exposure increases social play (Gemmel et al., 2017). Although perinatal exposure to SSRIs in relation to social play is less well studied, increases in serotonin due to the absence of the serotonin transporter or acute administration of fluoxetine has resulted in reductions in social play during adolescence, at least in males (Homberg et al 2007)(Kiser et al., 2012). Interestingly *perinatal* fluoxetine exposure has the opposite effect and it may be that altering the central serotonergic system with SSRIs during development alters neural circuitry related to social play phenotypes and contributes to increased social play in male rat offspring throughout life.

Pre-gestational stress also had enduring effects on social behaviors in adult male offspring resulting in significantly less social investigation and less time interacting with a novel same-sex conspecific. Therefore, both pSSRIs and pre-gestational maternal stress have enduring and behaviorally distinct effects on social behaviors in adult male offspring. Although these behavioral phenotypes have not been investigated in the clinical population, this work does suggest that there is a link between pSSRIs and/or maternal mental illness and specific behavioral outcomes. Regardless, this work is in agreement with clinical work showing that *both* pSSRIs and maternal mental illness can have enduring effects on social behaviors in children (Brown et al., 2017; Man et al., 2015; Mezzacappa et al., 2017; Oberlander and Zwaigenbaum, 2017). For example, maternal mood symptoms and pSSRIs are related to increased internalising behaviors in 3 and 4 year old children (Oberlander et al., 2010). pSSRIs have a history of affecting early behavioral development, even when controlling for maternal mental illness: Infants exposed to SSRIs during gestation exhibit blunted emotional and physical response to painful stimuli at two months of age, low adaptive and social-emotional behavioral scores at 10 months of age (Hanley et al., 2013; Oberlander et al., 2002; Oberlander et al., 2005), increased scores on the Pervasive Developmental Disorder scale at 2-5 years of age (Johnson et al., 2016), and increased externalizing behavior at 4 years of age (Hermansen et al., 2016; Oberlander et al., 2007). However, it must be acknowledged that the underlying state of maternal mental health and stress, as well as additional

environmental factors (Park and Oberlander, 2018), can mediate or moderate these pSSRI effects. Thus, the present work expands clinical work showing that pSSRIs and maternal depression may both contribute to developing social behaviors, particularly in male offspring.

That being said, findings from the present study show that pSSRIs and pre-gestational maternal stress have distinct effects on behavioral outcomes in adult male and female offspring. For example, pSSRIs increase social investigation in adult female offspring, while increasing social play in adult male offspring. Furthermore, pre-gestational maternal stress has little effect on social behaviors in female offspring, but decreases social investigation in adult male offspring. These sex differences may not be surprising as it is well documented in animal literature that social behaviors are sexually differentiated with male rodents, being more likely to engage in social play interactions while females being more likely to engage in social investigation and contact behaviors(Taylor et al., 2000). Therefore, pre-disposition toward specific behaviors, as well as natural variation in the frequency of male versus female social behaviors, may play a role in mediating the sex-dependent behavioral outcomes after pSSRIs or pre-gestational maternal stress.

While there are no definitive rodent behavioral tests which completely mimics human social behaviors, these findings show that perinatal, and pre-gestational factors, can have distinct and enduring patterns of effects on social behaviors that are sexually differentiated. Thus, further work on pSSRIs as treatment for maternal mood disorders should consider the role that fetal sex and child sex and/or gender may play on observed outcomes. In line with this, a recent meta-analysis addressing the relationship between pSSRIs and developmental disorders reported significant relationships with pSSRIs only when addressing male outcomes, or when male bias is included in the sample size (4:1 male to female ratios) (Harrington et al., 2014; Man et al., 2015). Therefore, human work associating pSSRIs to social behaviors needs further incorporation of both male and female offspring (Clements et al., 2015; El Marroun et al., 2014).

It is important to note that main effects of perinatal fluoxetine or pre-gestational maternal stress were evident in adult offspring but few, if any, interactions between the two factors existed. We have previously shown that pSSRIs can normalize neurobehavioral outcomes in juvenile and adolescent male and female offspring, but this is not the case in adulthood. For example, perinatal fluoxetine exposure prevents the effects of pre-gestational stress on sibling play in pre-adolescent offspring, but the same effect is not seen in adult offspring (Gemmel et al., 2017; Rayen et al., 2015). This suggests that pre-gestational maternal stress and pSSRIs have distinct neurobehavioral effects in adulthood, as discussed above; pSSRIs are less effective in ameliorating effects of maternal stress in the long term, particularly in male offspring; and gonadal hormones of puberty play a significant role in the long-term effects of pSSRIs and maternal stress, particularly in adult female offspring.

4.2. Perinatal SSRIs, Pre-gestational Stress, and HPA Outcomes

In the present study, perinatal fluoxetine affect GR density in the dentate gyrus of adult females and GR density in the CA3 of adult males. Perinatal SSRIs affect the serotonergic system (Gemmel et al., 2016a; Huang et al., 2012; Oberlander et al., 2009) and serotonin can act to increase the density of hippocampal glucocorticoid receptors (Erdeljan et al., 2001, 2005) (Meaney et al., 1994) (Laplante et al., 2002). Therefore, pSSRI exposure, which alter serotonin levels in the fetus, have long-term effects in modulating the density of hippocampal glucocorticoid receptors which likely affects a number of behavioral outcomes. In line with this we found a positive correlation between the GR density in the dentate gyrus and the time spent interacting with a novel same-sex conspecific in adult male, but not female, offspring. This suggests that HPA responsivity may be more precisely regulated as a function of increased GR density and thus make interacting with a novel partner less adversive, particularly in adult males after pSSRI exposure.

4.3. Perinatal SSRIs, Pre-gestational Stress, and Plasticity in the Hippocampus

In the present study we found a significant effect of perinatal fluoxetine exposure in reducing post-synaptic density in the CA2 region of the hippocampus in both adult male and female offspring. We

have also found that, during pre-adolscence, time interacting with a novel conspecific was significantly negatively correlated with the density of both pre- and post- synaptic proteins (Gemmel et al., 2017). Therefore, reductions in post synaptic density in the CA2 region of the hippocampus may play a role in increasing specific social interactions and, perhaps, social memory in adult offspring following pSSRI exposure.

When looking at patterns of hippocampal neurogenesis, we found that perinatal fluoxetine increased neurogenesis in the granule cell layer of adult female offspring. This finding supports and expands our previous work showing that early postnatal fluoxetine exposure increases new cell survival in adult female offspring (Rayen et al., 2015). In adult male offspring, there were no enduring effects of pSSRIs on hippocampal neurogenesis, but pre-gestational maternal stress reduced neurogenesis in the granule cell layer. Much research has shown that prenatal stress decreases hippocampal neurogenesis in adult male, but not female, offspring (Loi et al., 2014; Rayen et al., 2015), but this is the first to show that stress prior to gestation can have these enduring effects in adult male offspring. This also expands our previous work in pre-adolescent offspring showing that pre-gestational maternal stress, regardlesss of perinatal fluoxetine exposure, significantly reduced neurogenesis in the granule cell layer of males, but not females (Gemmel et al., 2017). Taken together this work suggests that neurogenesis in the hippocampal dentate gyrus may contribute to the sex-specific behavioral outcomes observed in the present study, with females being more sensitive to perinatal fluoxetine exposure and males being more sensitive to pre-gestational maternal stress.

4.4. Conclusions

The current work demonstrates clear effects of perinatal exposure to the SSRI fluoxetine on social investigation, social play, and neuroplasticity in adult male and female offspring. This works points to a need to consider the sex specific effects of perinatal SSRIs and maternal mood disorders on

neurobehavioral outcomes in children. Furthermore this work suggests that fetal-sex specific treatments, in general, may be warranted.

FIGURE LEGENDS

Figure 1. Representative photomicrographs of A) doublecortin in the GCL, B) synaptophysin-ir in the CA3, C) PSD-95-ir in the CA2, and D) GR-ir cells in the dentate gyrus of control female offspring at 40x objective. (*Scale bar* is set at 25 μm)

Figure 2. Mean (+SEM) time in social behaviors during social interaction in adult female and male offspring (s). A, D) time interacting with a stimulus animal; B, E) time in social investigation; C, F) time in social play. *denotes significance, ^ denotes significant main effect of treatment ($p<0.05$). CV = control + vehicle, CF= control + fluoxetine, PGSV= pre-gestational stress + vehicle, PGSF= pre-gestational stress + fluoxetine.

Figure 3. Measures of HPA outcomes. Mean (±SEM) GR density (OD) in the DG (A, D) and CA3 (B, E) of adult female and male offspring. C, F) Correlations between time in social interaction with a novel conspecific and GR density in the dentate gyrus of female and male offspring. *denotes significance, ^ denotes significant main effect of treatment ($p<0.05$). CV = control + vehicle, CF= control + fluoxetine, PGSV= pre-gestational stress + vehicle, PGSF= pre-gestational stress + fluoxetine. (n=6-16/sex/group)

Figure 4. Mean (±SEM) of (A, C) immature neurons (DCX-ir cells/slice) in the dorsal GCL and (B, D) PSD-95-ir in the CA2 region of the hippocampus. *denotes significance, ^ denotes significant main effect of treatment ($p<0.05$). CV = control + vehicle, CF= control + fluoxetine, PGSV= pre-gestational stress + vehicle, PGSF= pre-gestational stress + fluoxetine. (n=6/sex/group).

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Table 1. Chronic unpredictable stress (CUS) schedule prior to gestation. Females in the pre-gestational stress group were housed individually on day 0 and subjected to chronic unpredictable stress consisting of 0-2 stressors per day for 3 weeks. Previously published in Gemmel et al, 2017c.

Day of CUS	Stressor 1	Duration of Stressor 1	Stressor 2	Duration of stressor 2
1	Damp Bedding	8h day time	Overcrowding	24 hours
2	Restraint Stress	1 hour	Cage Rotation	12 hours
3	Overcrowding	24 hours	-	-
4	Food Depravation	8-12 hours	Swim Stress	5 minutes
5	Restraint Stress	1 hour	Food Depravation	12 hours
6	Swim Stress	5 minutes	Overcrowding	24 hours
7	Restraint Stress	1 hour	-	-
8	Damp Bedding	8 hours	Cage Rotation	12 hours
9	Restraint Stress	1 hour	Food Depravation	12 hours
10	Damp Bedding	8 hours	Overcrowding	24 hours
11	-	-	-	-
12	Cage Rotation	12 hours	Swim Stress	5 minutes
13	Restraint Stress	1 hour	Swim Stress	5 minutes
14	Damp Bedding	8 hours	Overcrowding	24 hours
15	Restraint Stress	1 hour	Cage Rotation	12 hours
16	Food Depravation	12 hours	Overcrowding	24 hours
17	Restraint Stress	1 hour	Restraint Stress	1 hour
18	Overcrowding	24 hours	-	-
19	Swim Stress	5 minutes	Swim Stress	5 minutes
20	Cage Rotation	1 hour	Damp Bedding	8 hours
21	Overcrowding	24 hours	Food Depravation	12 hours

Table 2. Significant three-way ANOVA main effects with Condition (pre-gestational stress/control), Treatment (fluoxetine/vehicle) and Sex (female/male) on measures of social behaviors, HPA axis, and synaptic outcomes.

	Main Effect of Condition	**Main Effect of Treatment**	**Main Effect of Sex**
Social Behavior			
	sniffing (F(1,79)=15.2, p=0.0002) crawling (F(1,79)=3.97, p=0.049) social investigation (F(1,79)=15.67, p=0.0002) all scored behavior (F(1,79)=5.89, p=0.02)	social investigation (F(1,79)=6.64, p=0.01) all scored behavior (F(1,79)=4.57, p=0.04).	sniffing (F(1,79)=7.10, p=0.009) social investigation (F(1,79)=5.16, p=0.03)
HPA			
	Corticosterone (F(1,74)=5.33, p=0.02) CBG (F(1,79)=6.97, p=0.01)		free corticosterone (F(1,74)=68.26, p<0.00001) Corticosterone (F(1,74)=187.52, p<0.00001) CBG (F(1,79)=678.48, p<0.00001), GR density in DG (F(1,40)=12.84, p=0.0009) GR density in CA3 (F(1,40)=10.26, p=0.003)
Hippocampus			
	DCX-ir cells (F(1,40)=8.54, p=0.006).	PSD-95-ir in CA2 (F(1,40)=11.57, p=0.002)	

Table 3. Mean (±SEM) time to first interaction and duration (s) in scored behaviors with a novel stimulus conspecific. *denotes significance, ^denotes significant main effect of treatment ($p<0.05$). (n=8-16/sex/group)

	CV	**CF**	**PGSV**	**PGSF**
	Female			
Time to first interaction	13.6 ± 2.6	12.4 ± 2.1^	19.8 ± 3.5	6.1 ± 1.6^
Sniffing	86.5 ± 6.5	114.3 ± 7.8^	70.0 ± 8.6*	94.4 ± 8.1^*
Following	32.5 ± 3.4	48.2 ± 8.2^	26.3 ± 6.0	50.6 ± 4.7^
Crawling	55.0 ± 6.8	48.2 ± 6.8	58.8 ± 7.3	59.4 ± 6.4
Grooming partner	6.0 ± 1.8	7.9 ± 1.5^	2.5 ± 0.9	8.3 ± 2.5^
Running Away	10.0 ± 2.8	14.3 ± 2.7	18.1 ± 4.4	11.1 ± 1.4
Self-grooming	16.8 ± 4.9	8.9 ± 1.4	15.9 ± 3.0	14.9 ± 4.1
	Male			
Time to first interaction	9.2 ± 1.7	9.5 ± 1.3	10.4 ± 3.1	18.2 ± 4.8
Sniffing	123.8 ± 6.7	115.9 ± 7.2	93.1 ± 8.1*	93.5 ± 8.9*
Following	50.4 ± 4.9	57.8 ± 4.9	35.0 ± 4.8*	31.0 ± 5.8*
Crawling	45.4 ± 5.1	42.8 ± 4.4	59.4 ± 6.4	50.0 ± 7.0
Grooming partner	10.8 ± 1.9	9.7 ± 2.0	6.3 ± 2.3	7.0 ± 1.7
Running Away	7.1 ± 2.2	11.9 ± 1.9^	5.0 ± 1.3	16.0 ± 3.5^
Self-grooming	9.1 ± 2.3	13.2 ± 2.3^	8.5 ± 1.8	15.8 ± 2.5^

Table 4. Mean (±SEM) serum corticosterone (nM), CBG (nM) and free corticosterone (n=8-16/group), and glucocorticoid receptor (GR) density (OD) in the hippocampus and PVN (n=5-6/group). * denotes significant main effect of pre-gestational stress ($p<0.05$). CV = control + vehicle, CF= control + fluoxetine, PGSV= pre-gestational stress + vehicle, PGSF= pre-gestational stress + fluoxetine.

	CV	**CF**	**PGSV**	**PGSF**
	Female			
Corticosterone (nM)	1974.7±348.47	2099.4±192.9	1726.0±188.4*	1383.6±153.1*
CBG (nM)	1980.1± 127.3	2023.4±58.4	1654.3 ± 84.5*	1931.4± 70.7*
Free corticosterone (nM)	100.6 ± 17.6	101.9 ± 8.9	105.8 ± 13.1	73.8 ± 9.7
GR-ir CA2	0.10 ± 0.01	0.10 ± 0.02	0.08 ± 0.01	0.09 ± 0.01
GR-ir PVN	0.23 ± 0.03	0.18 ± 0.01	0.17 ± 0.01	0.18 ± 0.04
	Male			
Corticosterone (nM)	226.6 ± 41.0	284.7 ± 63.6	279.4 ± 51.4	143.0 ± 48.1
CBG (nM)	765.0±19.2	746.0±26.1	733.7±29.8	726.9±31.5
Free corticosterone (nM)	29.7 ± 5.4	41.1 ± 10.0	39.8 ± 8.3	19.6 ± 6.6
GR-ir CA2	0.07 ± 0.004	0.07 ± 0.01	0.08 ± 0.01	0.08 ± 0.01
GR-ir PVN	0.17 ± 0.03	0.21 ± 0.02	0.16 ± 0.01	0.18 ± 0.01

Table 5. Mean (±SEM) synaptic protein density (OD) in the hippocampus (n=6/group). CV = control + vehicle, CF= control + fluoxetine, PGSV= pre-gestational stress + vehicle, PGSF= pre-gestational stress + fluoxetine.

	CV	**CF**	**PGSV**	**PGSF**
	Female			
Synaptophysin				
DG	0.19 ± 0.03	0.16 ± 0.02	0.15 ± 0.01	0.20 ± 0.03
CA2	0.05 ± 0.02	0.07 ± 0.03	0.02 ± 0.01	0.07 ± 0.02
CA3	0.16 ± 0.03	0.13 ± 0.01	0.11 ± 0.01	0.15 ± 0.02
PSD-95				
DG	0.04 ± 0.007	0.05 ± 0.004	0.04 ± 0.006	0.04 ± 0.005
CA3	0.05 ± 0.007	0.04 ± 0.005	0.05 ± 0.006	0.04 ± 0.004
	Male			
Synaptophysin				
DG	0.22 ± 0.02	0.18 ± 0.02	0.18 ± 0.03	0.21 ± 0.03
CA2	0.07 ± 0.01	0.07 ± 0.01	0.07 ± 0.02	0.10 ± 0.01
CA3	0.16 ± 0.01	0.13 ± 0.01	0.14 ± 0.03	0.15 ± 0.02
PSD-95				
DG	0.06 ± 0.01	0.04 ± 0.004	0.04 ± 0.006	0.03 ± 0.004
CA3	0.05 ± 0.009	0.04 ± 0.003	0.05 ± 0.006	0.06 ± 0.03

Figure 1

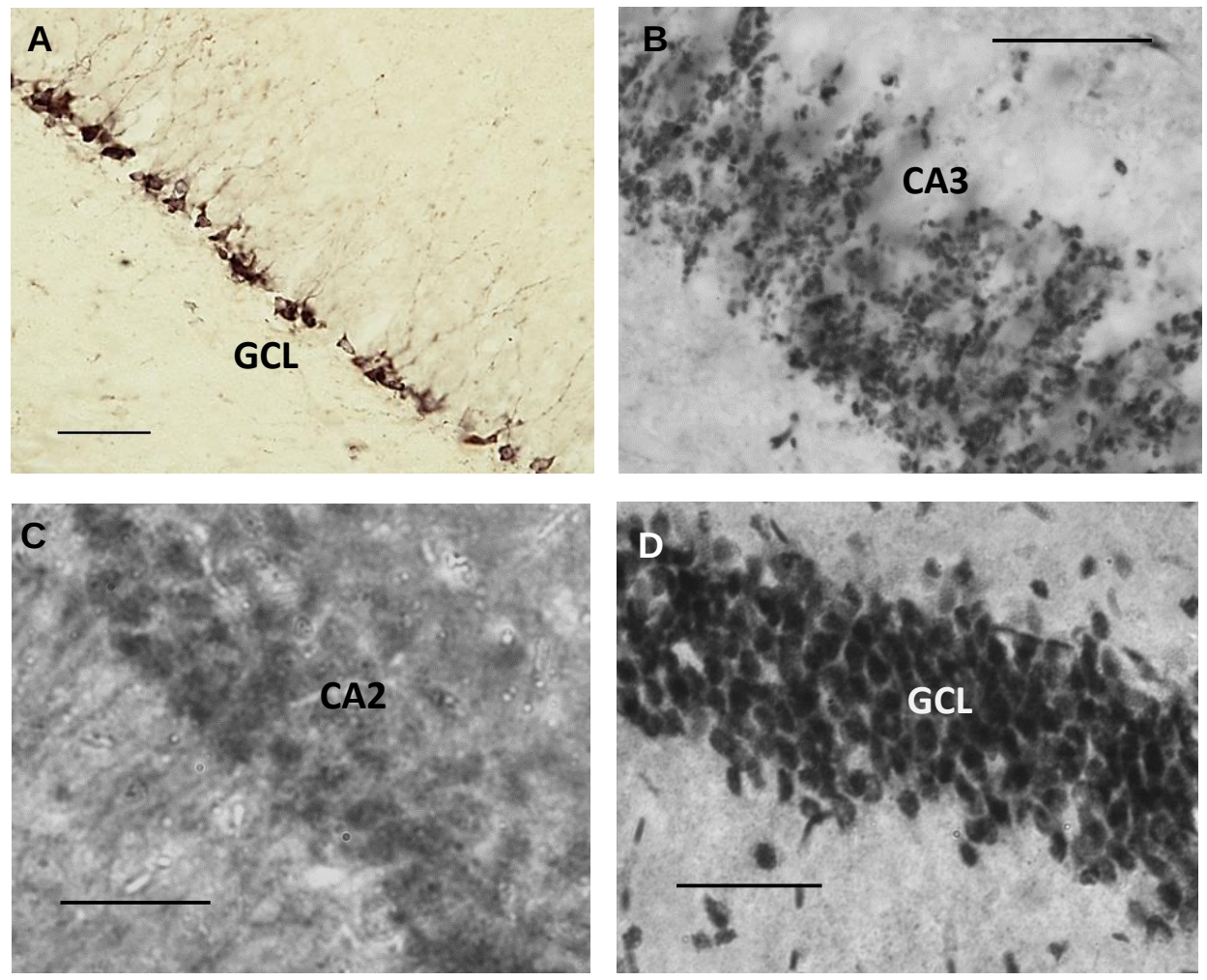
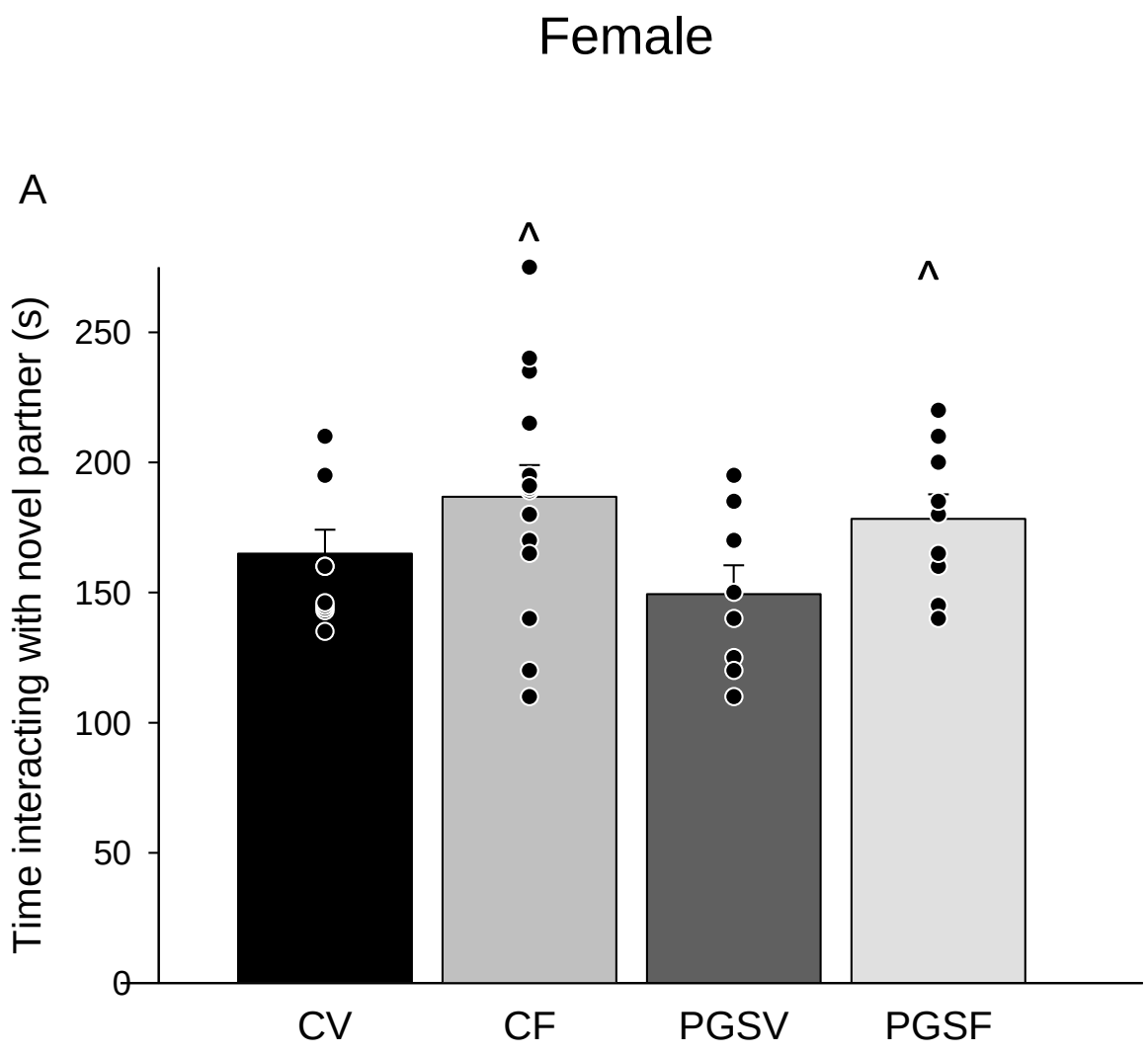
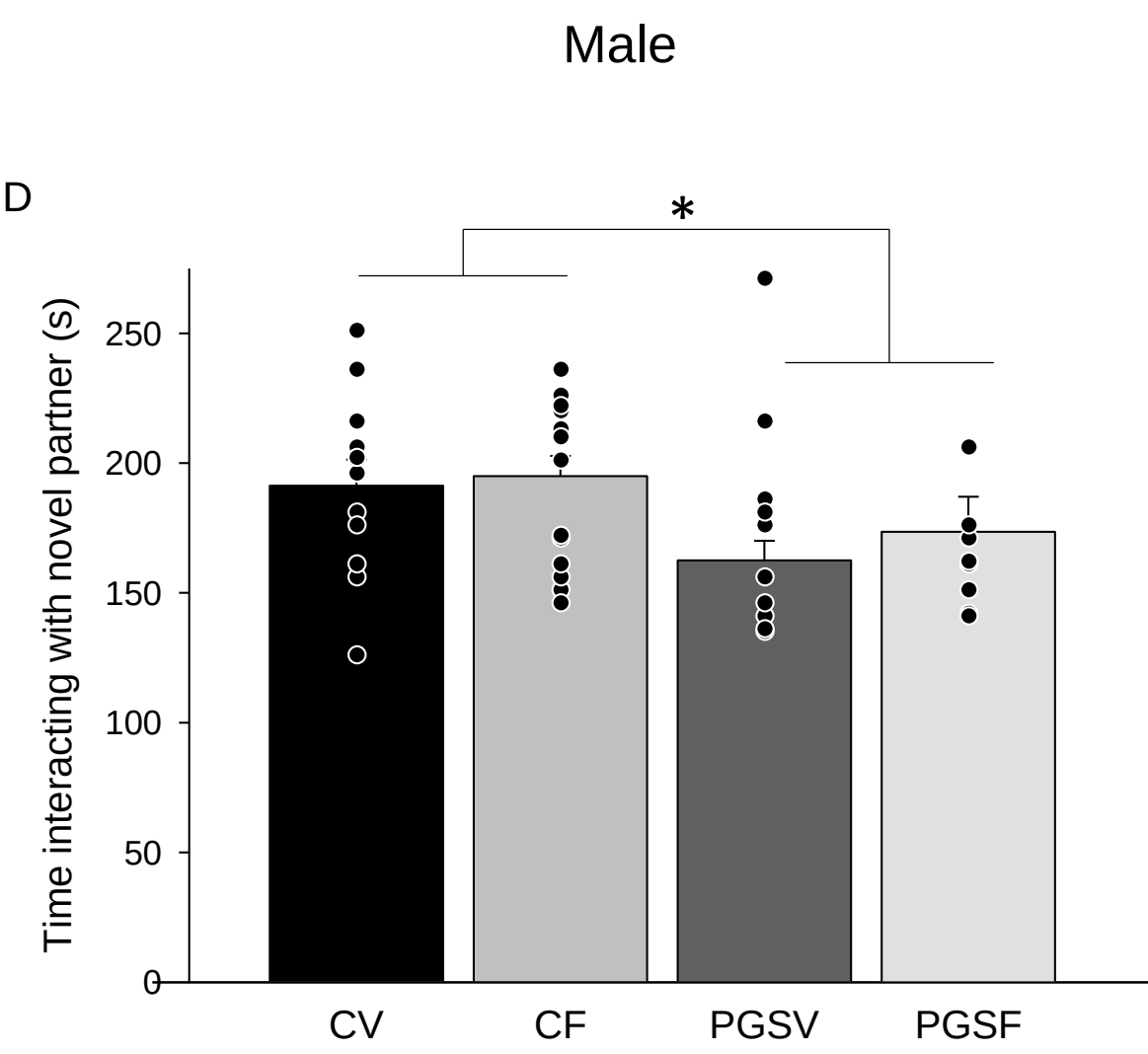
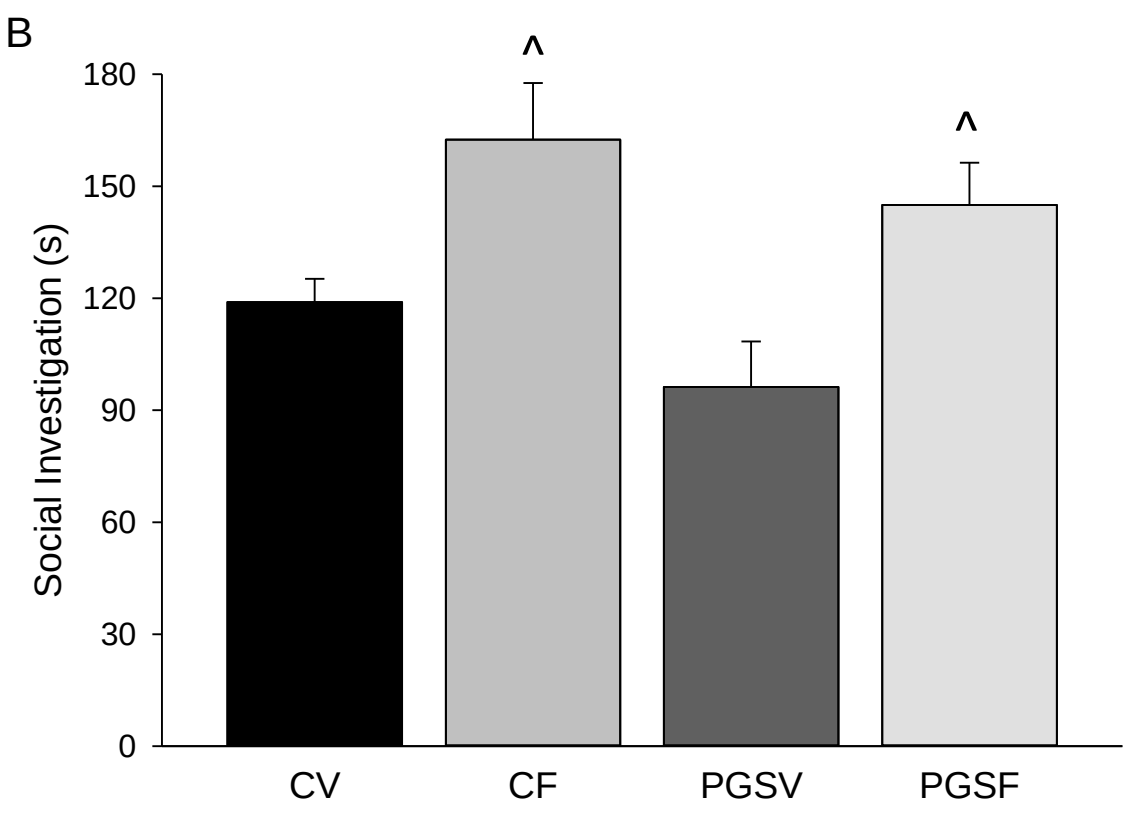
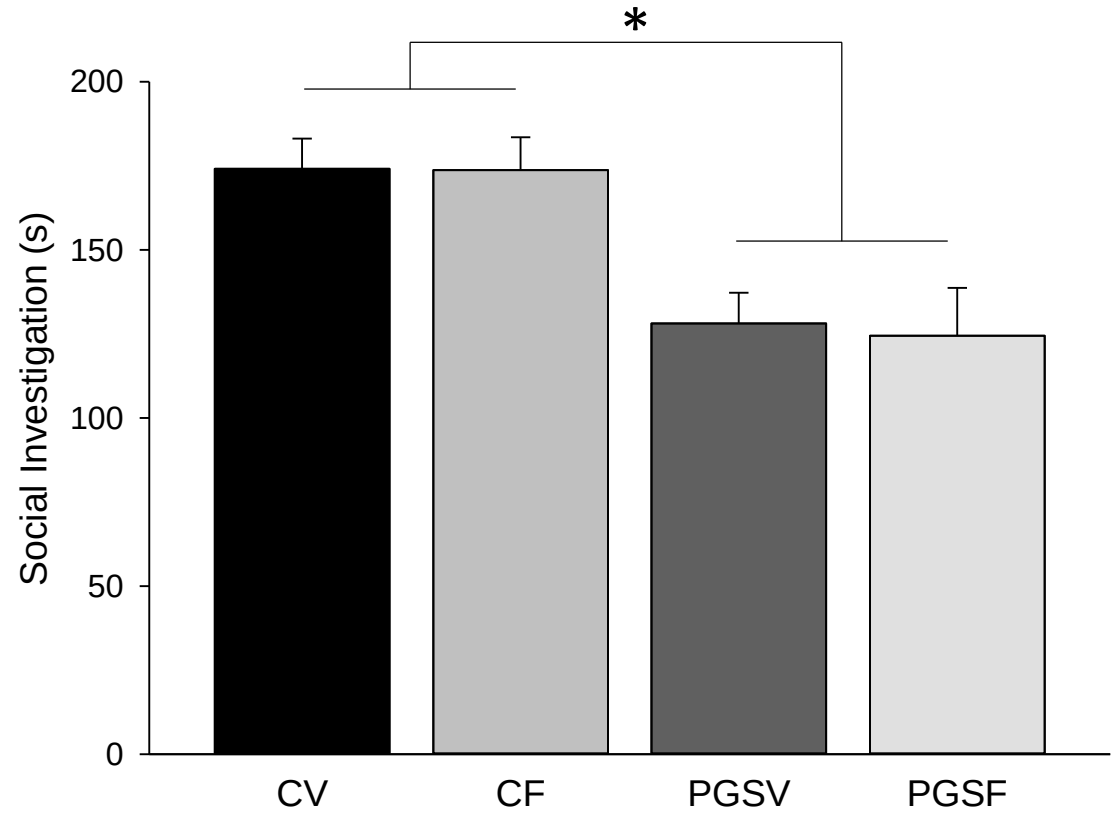


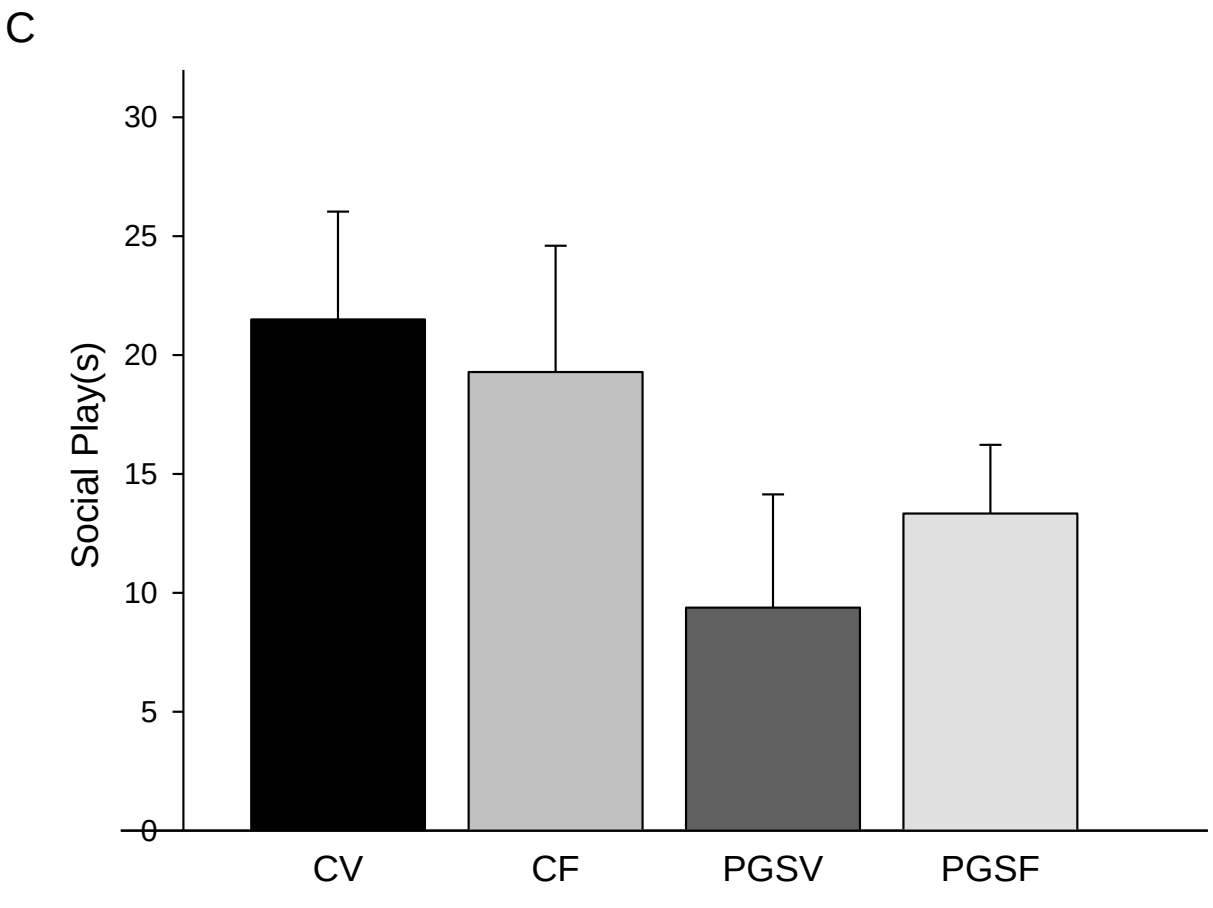
Figure 2


Female
A
Time interacting with novel partner (s)
0
50
100
150
200
250
^
^
CV
CF
PGSV
PGSF


Male
D
Time interacting with novel partner (s)
0
50
100
150
200
250
*
CV
CF
PGSV
PGSF


B
Social Investigation (s)
0
30
60
90
120
150
180
^
^
CV
CF
PGSV
PGSF


E
Social Investigation (s)
0
50
100
150
200
*
CV
CF
PGSV
PGSF


C
Social Play(s)
0
5
10
15
20
25
30
CV
CF
PGSV
PGSF

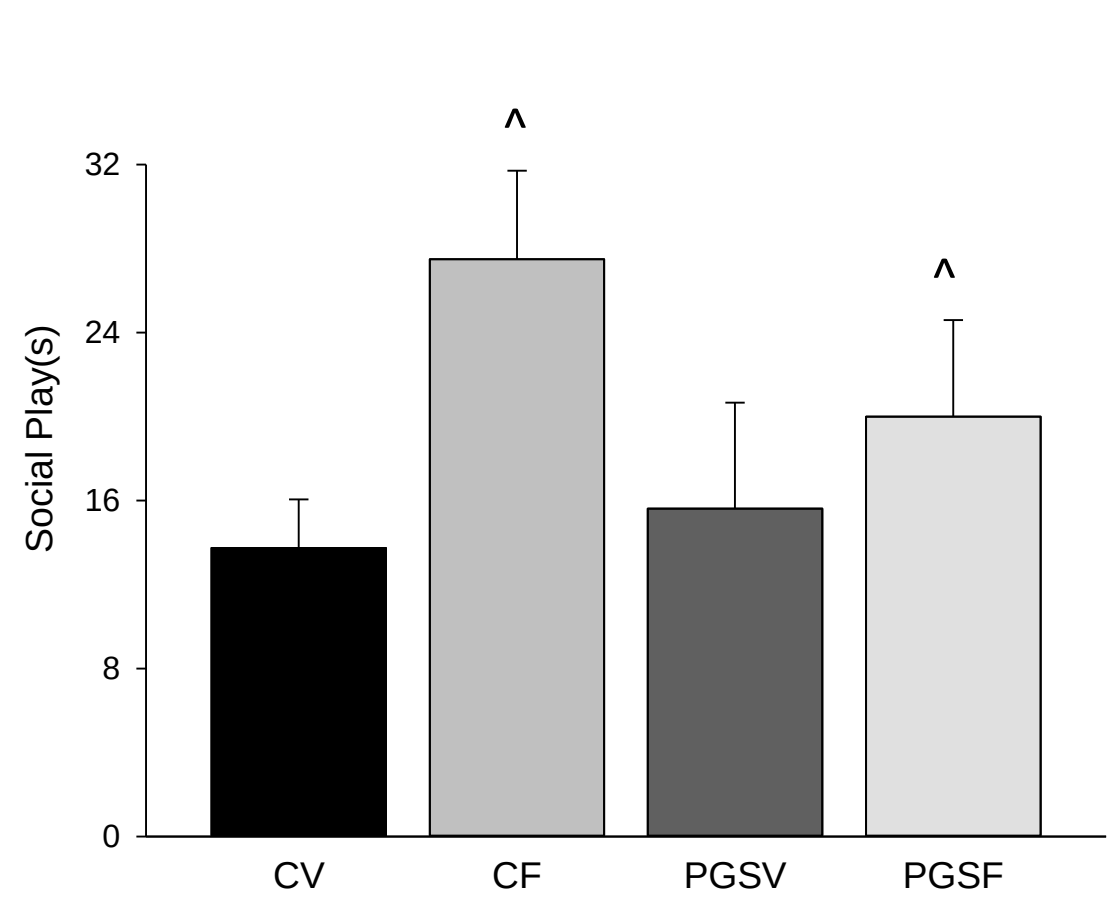
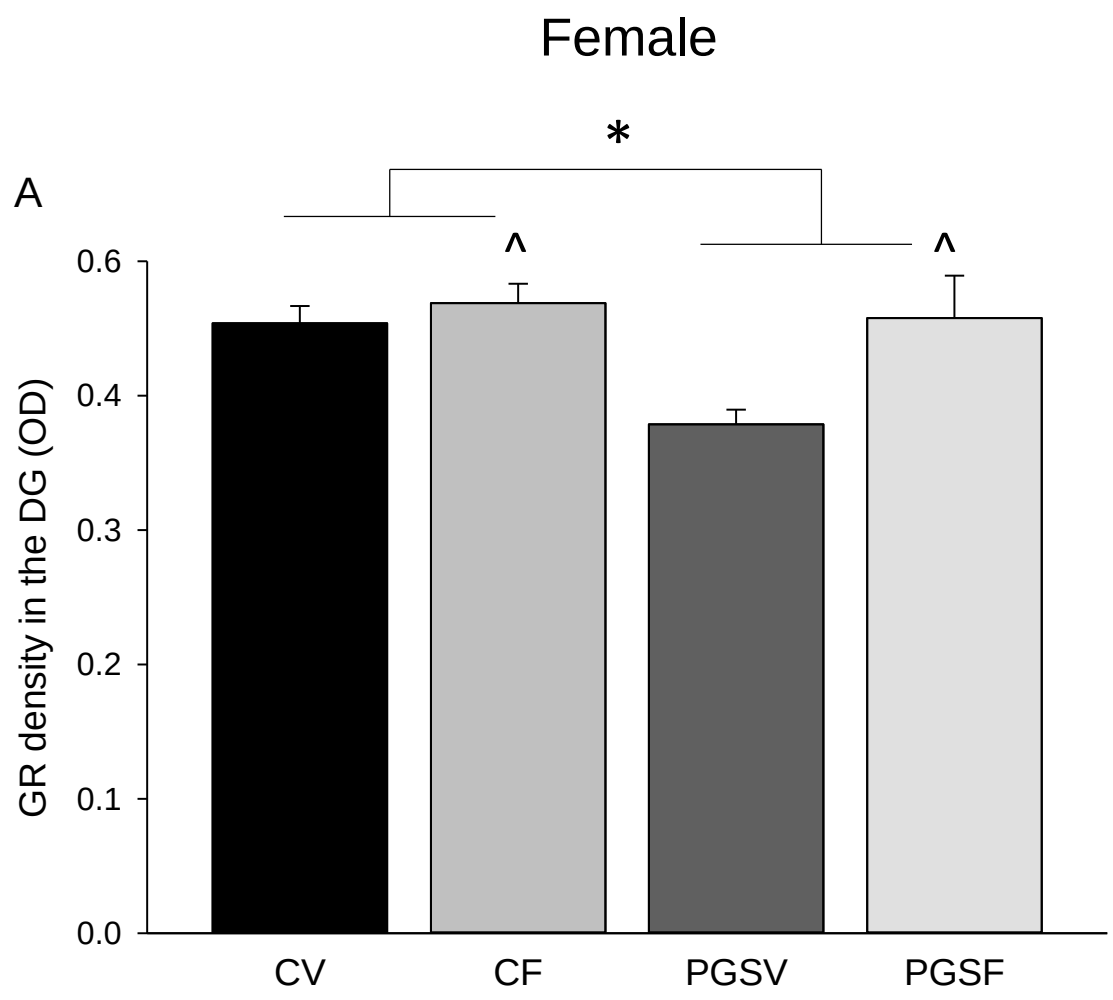
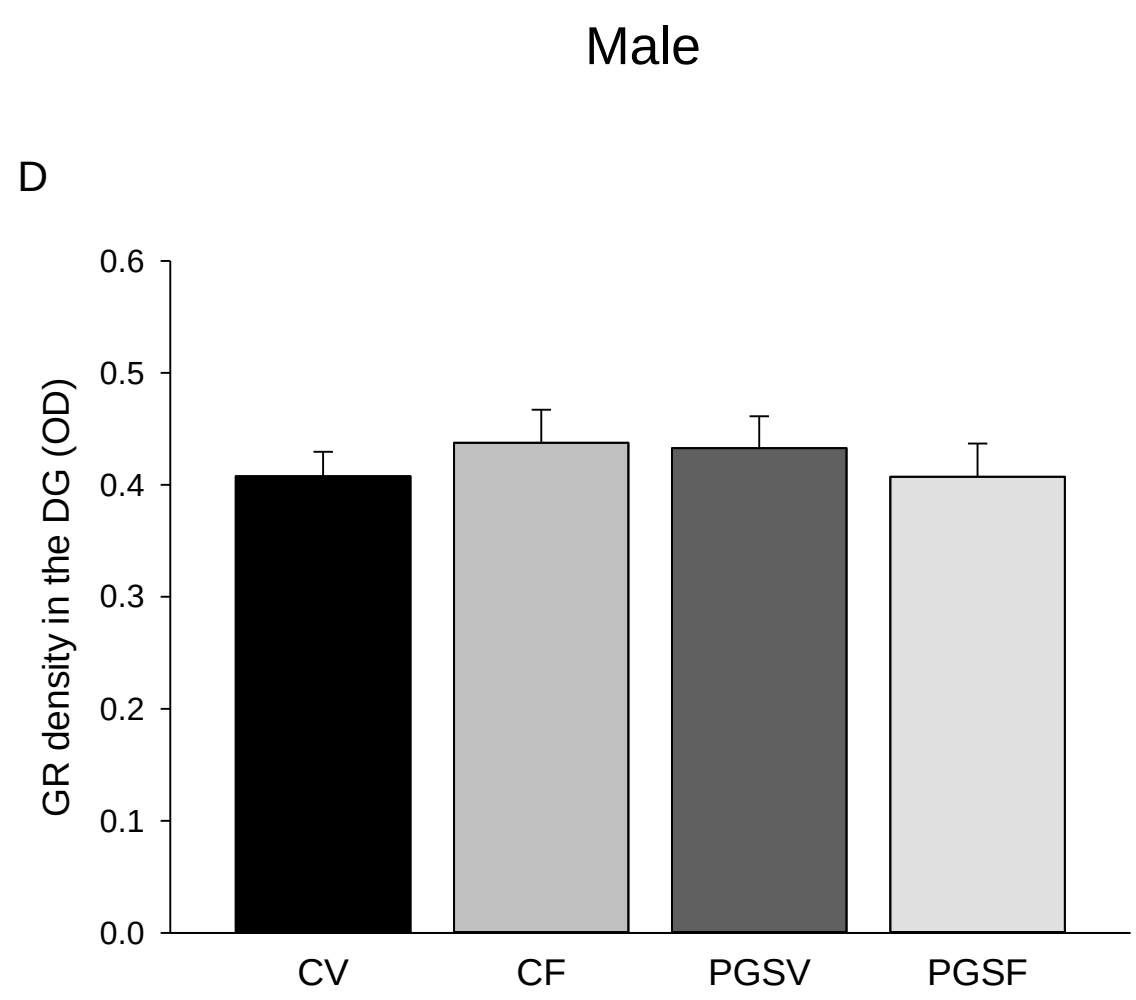
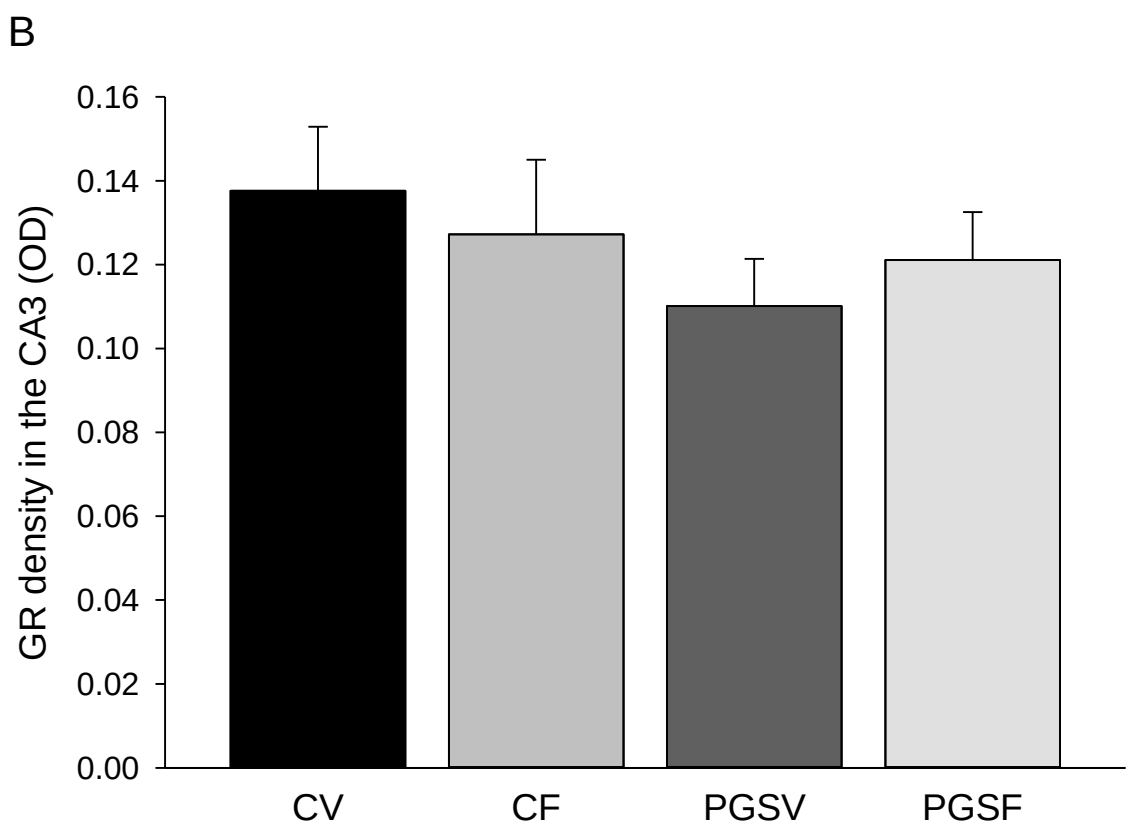
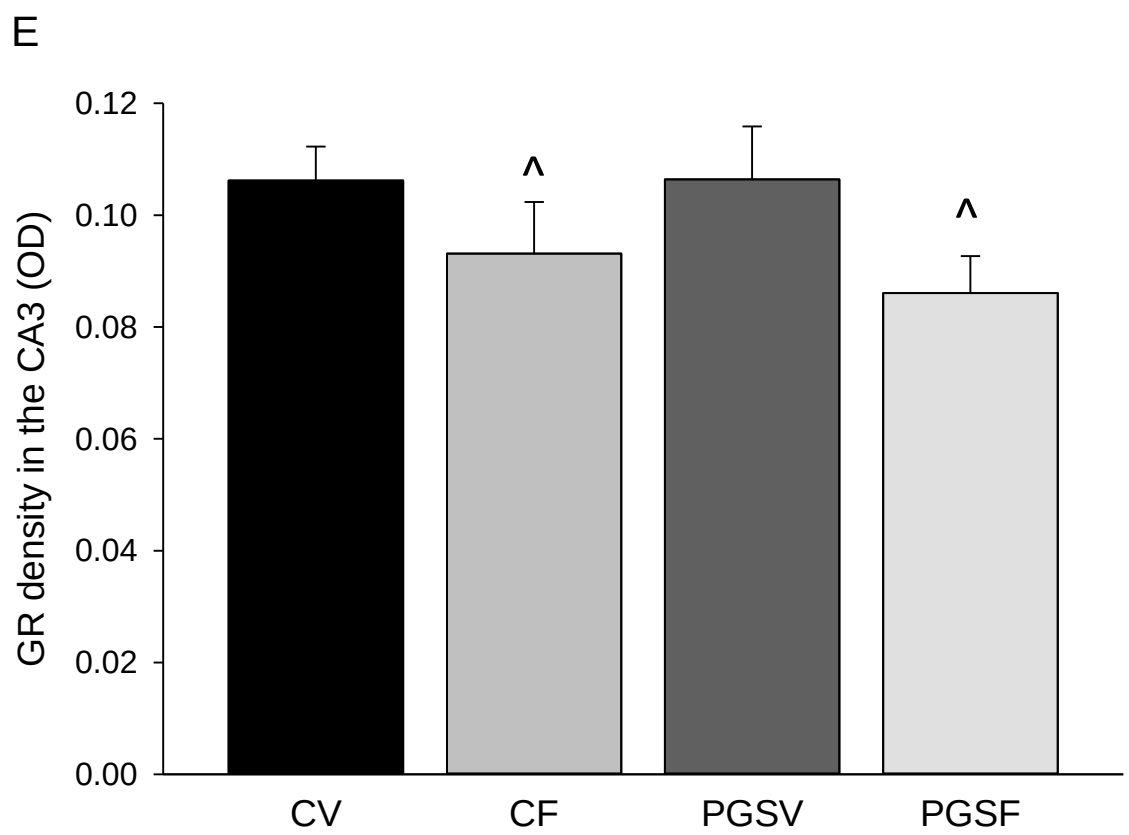

F
Social Play(s)
0
8
16
24
32
^
^
CV
CF
PGSV
PGSF

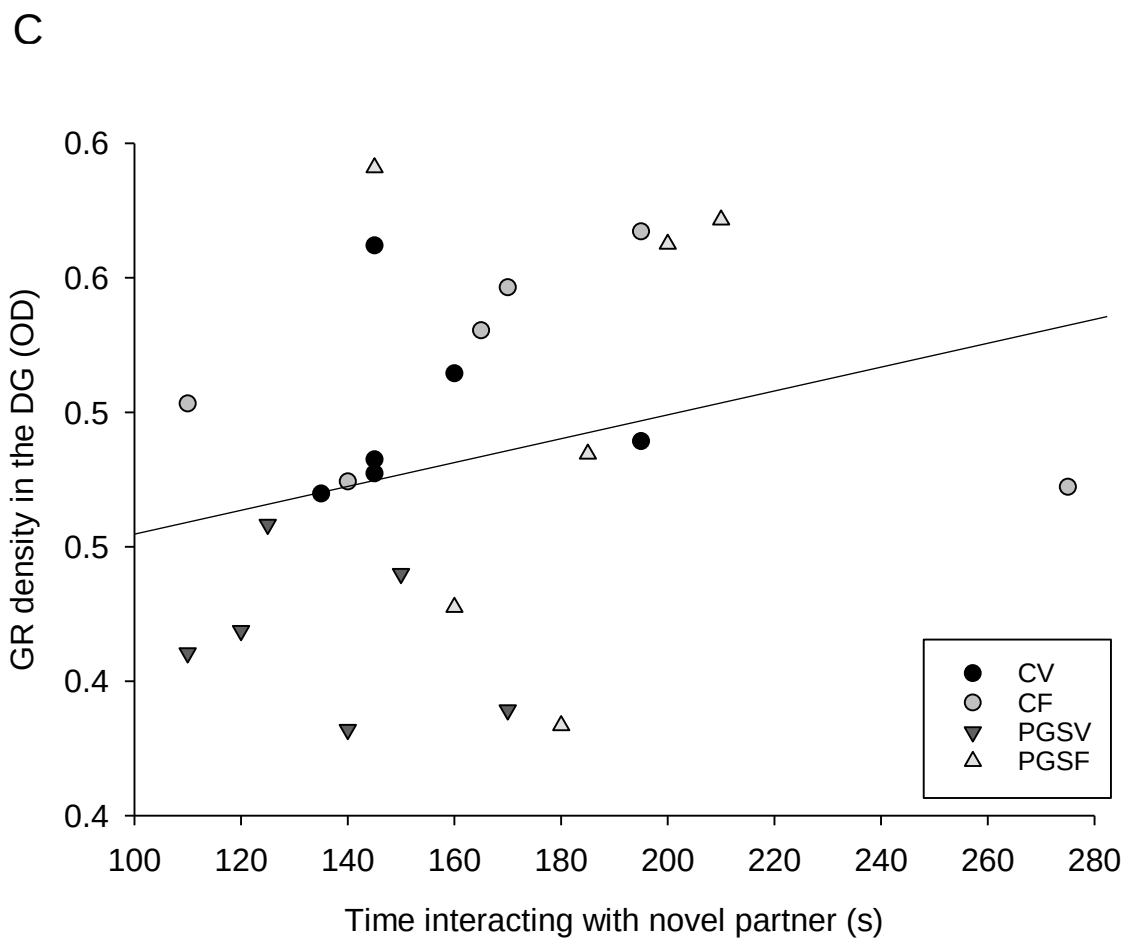
Figure 3


Female
A
*
^
^
GR density in the DG (OD)
0.6
0.4
0.3
0.2
0.1
0.0
CV
CF
PGSV
PGSF


Male
D
GR density in the DG (OD)
0.6
0.5
0.4
0.3
0.2
0.1
0.0
CV
CF
PGSV
PGSF


B
GR density in the CA3 (OD)
0.16
0.14
0.12
0.10
0.08
0.06
0.04
0.02
0.00
CV
CF
PGSV
PGSF


E
GR density in the CA3 (OD)
0.12
0.10
0.08
0.06
0.04
0.02
0.00
^
^
CV
CF
PGSV
PGSF


C
GR density in the DG (OD)
0.6
0.6
0.5
0.4
0.4
100
120
140
160
180
200
220
240
260
280
Time interacting with novel partner (s)
CV
CF
PGSV
PGSF

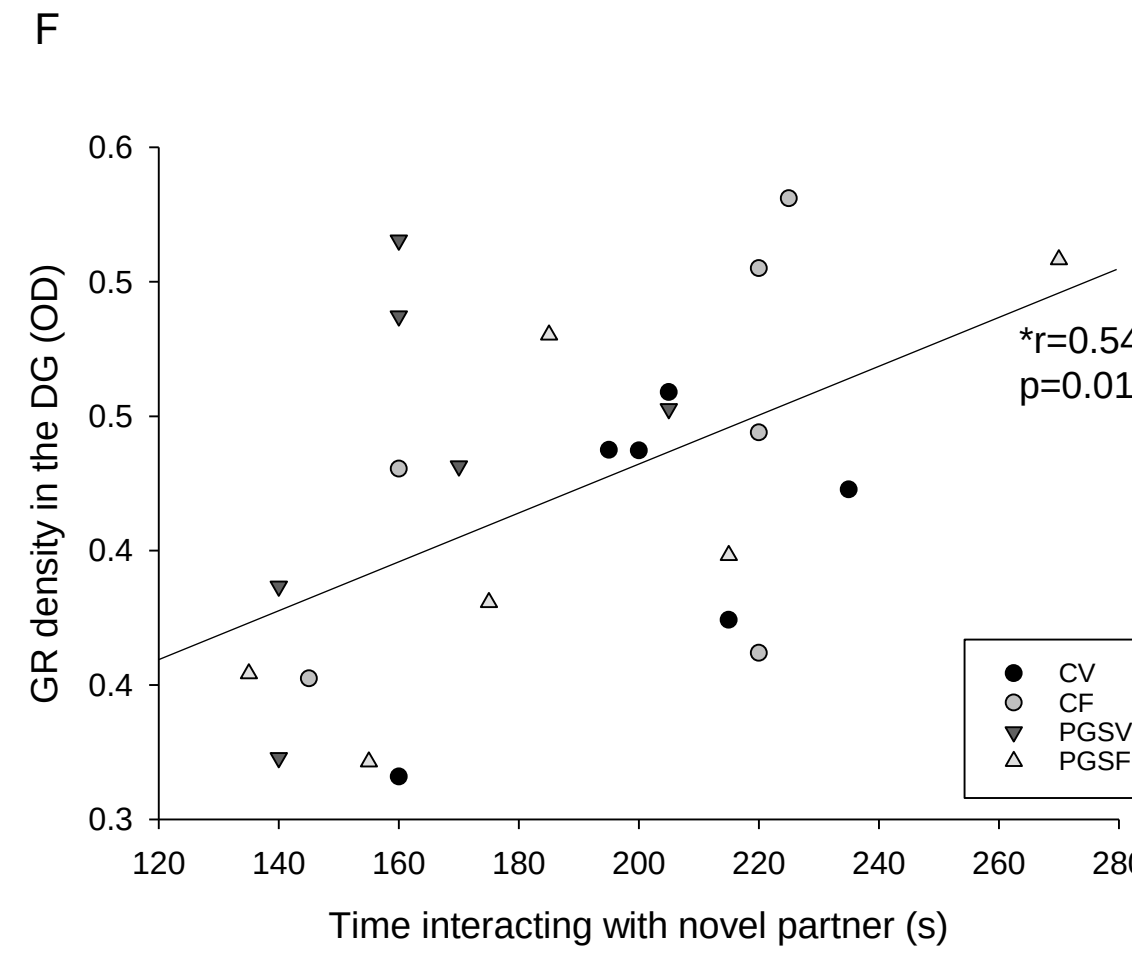
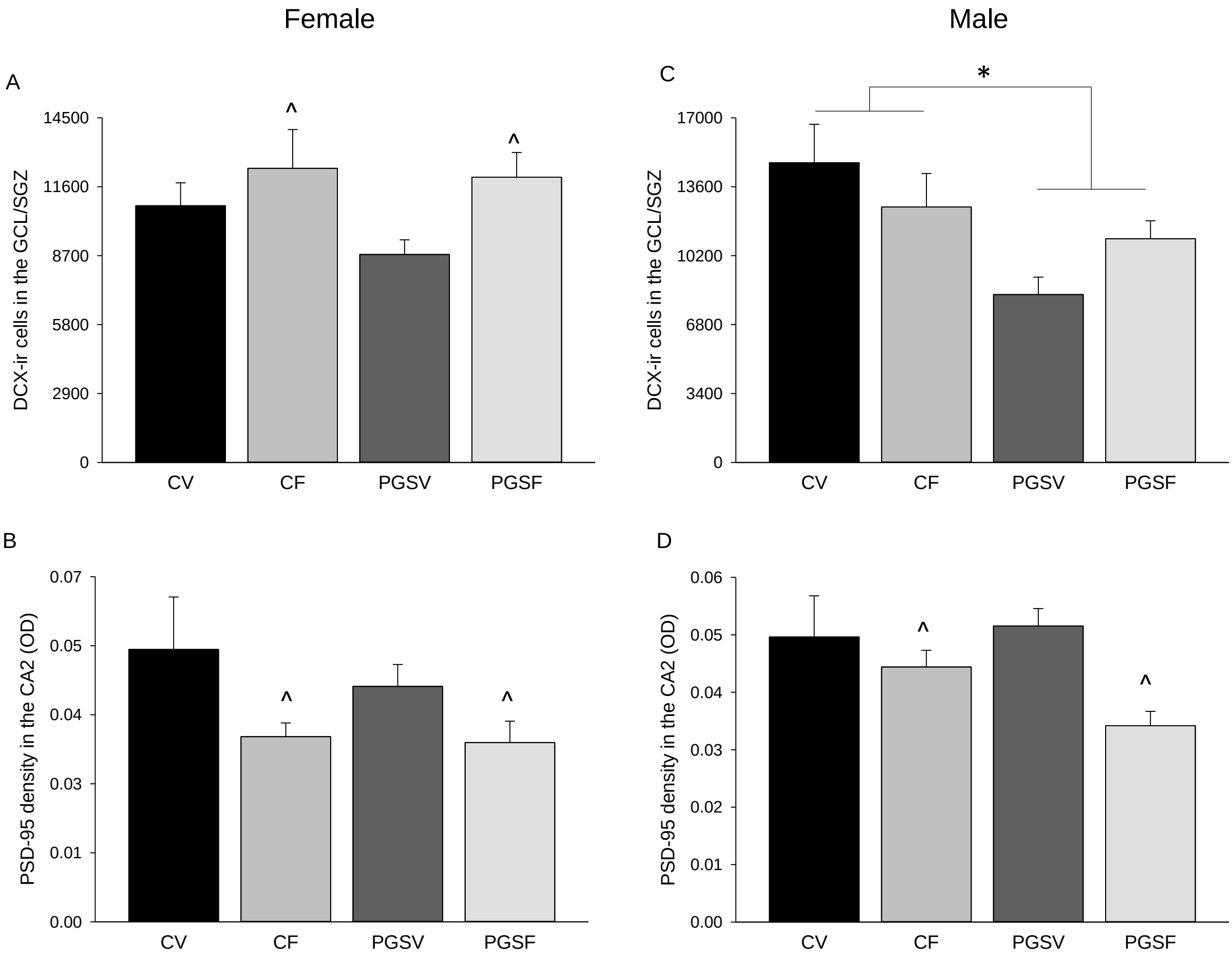

F
GR density in the DG (OD)
0.6
0.5
0.5
0.4
0.4
0.3
120
140
160
180
200
220
240
260
280
Time interacting with novel partner (s)
*r=0.54,
p=0.01
CV
CF
PGSV
PGSF

Figure 4


Female
Male
A
C
B
D
DCX-ir cells in the GCL/SGZ
PSD-95 density in the CA2 (OD)
14500
11600
8700
5800
2900
0
17000
13600
10200
6800
3400
0.07
0.05
0.04
0.03
0.01
0.00
0.06
0.02
^
*
CV
CF
PGSV
PGSF

Highlights

- Perinatal SSRIs increase social investigation in females and social play in males
- Pre-gestational maternal stress affects social investigation in adult males only
- Perinatal SSRIs have sexually differentiated effects on hippocampal plasticity
- Perinatal SSRIs and maternal stress affect GR density in the hippocampus
- Fetal-sex specific treatment should be considered for maternal mental illness