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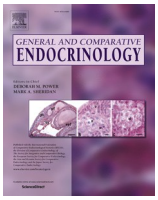

Acute intranasal oxytocin dose enhances social preference for parents over peers in male but not female peri-adolescent California mice (*Peromyscus californicus*)

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ABSTRACT

Peri-adolescence is a critical developmental stage marked by profound changes in the valence of social interactions with parents and peers. We hypothesized that the oxytocin (OXT) and vasopressin (AVP) systems, known for influencing social behavior, would be involved in the maintenance and breaking of bonding behavior expressed by very early peri-adolescent males and females. In rodents, OXT is associated with mother–pup bonding and may promote social attachment to members of the natal territory. AVP, on the other hand, can act in contrasting ways to OXT and has been associated with aggression and territoriality. Specifically, we predicted that in peri-adolescent male and female juveniles of the biparental and territorial California mouse (*Peromyscus californicus*), a) OXT would increase the social preferences for the parents over unfamiliar age-matched peers (one male and one female), and b) AVP would break the parent-offspring bond and either increase time in the neutral chamber and/or approach to their unfamiliar and novel peers. We examined anxiety and exploratory behavior using an elevated plus maze and a novel object task as a control. Peri-adolescent mice were administered an acute intranasal (IN) treatment of 0.5 IU/kg IN AVP, 0.5 IU/kg IN OXT, or saline control; five minutes later, the behavioral tests were conducted. As predicted, we found that IN OXT enhanced social preference for parents; however, this was only in male and not female peri-adolescent mice. IN AVP did not influence social preference in either sex. These effects appear specific to social behavior and not anxiety, as neither IN OXT nor AVP influenced behavior during the elevated plus maze or novel object tasks. To our knowledge, this is the first evidence indicating that OXT may play a role in promoting peri-adolescent social preferences for parents and delaying weaning in males.

1. Introduction

Across a wide variety of species, the developmental stage of adolescence is highly conserved (Spear, 2007). Mammals with shorter lifespans and less complex social environments enter puberty more quickly (Brenhouse & Andersen, 2011). For these species, the cost of prolonging time in their natal territory and competing with their siblings and parents for resources outweighs the benefits of parental food resources, protection, and social learning. However, a longer buffer period between puberty and adulthood may be advantageous for some species. For example, in species with dominance hierarchies, prolonged adolescence likely helps to attain higher social ranks and greater mating opportunities (Steinberg, 2010; Duell & Steinberg, 2019). Resource availability and group social composition may change across births from the same parent or across generations as areas experience drought or other cyclic environmental pressures (Prugh et al., 2018). Plasticity in weaning time could afford adolescents more time to prepare to explore social challenges in and out of their natal territory, migrate, and establish new territories when conditions are less favorable.

While environmental pressures guide a peri-adolescent mammal's weaning time, their social bond with their parent(s) may also be a proximate mechanism influencing weaning time. Among several candidate neurochemicals, there are two groups of likely hormonal mechanisms that could affect this process: one is the sex steroid hormones that influence maturation and the process of puberty (Romeo, 2003; Forbes & Dahl, 2010; Delevich et al., 2021) and the second is

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oxytocin (OXT) that influences bonding (Carter et al., 1992; Kendrick, 2000; Liu & Wang, 2003; Nagasawa et al., 2012; Loth & Donaldson, 2021), but is also influenced by those sex steroid hormones (Shapiro et al., 2000; Murakami et al., 2011). Dramatic steroid hormone changes are associated with the transition from being a juvenile to becoming an adult (Peper et al., 2011; Koolschijn et al., 2014; Trova et al., 2021), and these hormones can also drive the regulation of OXT and a similar neuropeptide, vasopressin (AVP) (De Vries et al., 1994). OXT and AVP influence social behavior and preference across a wide variety of mammalian species, including rodents (Kent et al., 2013; Lukas & Neumann, 2014; Wood et al., 2015; Duque-Wilckens et al., 2018; Williams et al., 2020; Bester-Meredith & Marler, 2001; Guoynes et al., 2018; Lukas et al., 2011; Albers, 2012; Winslow et al., 1993; Wang et al., 1999; Keverne & Curley, 2004; Prounis et al., 2018; Albers and Bamshad, 1999; Taylor et al., 2022; Taylor et al., 2017), and nonhuman and human primates (Taylor & French, 2015; Jarcho et al., 2011; Baxter et al., 2020; Crockford et al., 2013; Staes et al., 2015; Heinrichs et al., 2009; Skuse et al., 2014; Maud et al., 2018; Bredewold and Veenema, 2018; Zhuang et al., 2021). Here we focus on the OXT and AVP systems as neurochemical candidates for influencing weaning time in mammals. In mothers, the physiological release of oxytocin (OXT) promotes milk letdown (Nishimori et al., 1996; Young et al., 1997; Keverne and Kendrick, 1992), increases circulating OXT via increased pup suckling (Febo et al., 2005; Neumann et al., 1993), and changes OXTR receptor expression in social brains areas such as the nucleus accumbens, ventral tegmental area, bed stria of the nucleus terminalis, ventromedial hypothalamus, lateral septum (Keebaugh et al., 2015; Insel, 1990; Pedersen et al., 1994; Curley et al., 2012). In the early stages of rearing, a single intranasal dose of OXT is enough to promote increased maternal communication and care in California mice (Guoynes & Marler, 2021). These studies suggest a link between the OXT-driven positive feedback mechanisms associated with milk let-down/suckling and the mother-offspring bond.

Despite the above research on the effect of OXT on bond initiation in mothers, the neurobiological mechanisms that maintain the parent-offspring bond through peri-adolescence are not well understood. Is the bond maintained by the parent(s), the offspring, or both? Here we initially address this question through a mechanistic approach; we ask whether OXT might induce flexibility in weaning time. In young female and male rats, post-weaning isolation decreases OXT receptor expression in the nucleus accumbens and increases aggression, suggesting that higher OXT receptivity in the nucleus accumbens may be important for prosocial behavior (de Moura Oliveira et al., 2019). When offspring transition from the juvenile period to adulthood, OXTR expression decreases in the lateral septum, an area known for its role in integrating sensory information and prior social experiences (Smith et al., 2017). Therefore, it is possible that alterations to the OXT system not only facilitate changes to the mother's social preference and behavior but also in her offspring's preferences and behavior. Although our focus is on OXT and AVP, we also acknowledge the close connections between the sex steroid hormones and the OXT system (McCarthy et al., 1996; Frayne & Nicholson, 1995; Young et al., 1998; Murakami et al., 2011; Acevedo-Rodriguez et al., 2015) that underscore the possibility that the oxytocin system will affect females and males differently.

To test the hypothesis that OXT influences offspring-parent attachment during very early peri-adolescence, we use California mice (*Peromyscus californicus*), a monogamous, biparental rodent species whose social behavior can be modulated by OXT manipulations (Duque-Wilckens et al., 2018; Perea-Rodriguez et al., 2015; Guoynes & Marler, 2021; Monari et al., 2021; Guoynes & Marler, 2022; Gubernick et al., 1995; Williams et al., 2020; Steinman et al., 2019; Yohn et al., 2018). Like other species, the young mature and migrate from their natal territory. There is no published evidence of overlapping generations, and females and males disperse from the nest (Ribble, 1992). Interestingly, females disperse farther from their natal territory than males (Ribble, 1992), suggesting male juveniles may be less inclined to explore and show greater parent-offspring proximity and attachment behavior. In addition to testing the effect of OXT on parent-offspring bonding, we also wanted to examine whether AVP influenced parent-offspring bonding due to its role in territoriality and aggression in California mice (Bester-Meredith & Marler, 2001; Frazier et al., 2006; Yohn et al., 2017; Bester-Meredith et al., 1999). We predicted that while OXT would maintain the parent-offspring bond, AVP may erode the parent-offspring bond by increasing territoriality in the peri-adolescents. Moreover, we expected that if any sex showed a stronger or more flexible bond with the parents, it would be males because of staying closer to the natal territory. We tested these predictions using a three-chambered choice test in which peri-adolescents, close to weaning age, chose between spending time with two sets of social partners: their parents versus an unrelated, age-matched novel female and male. As a control, we wanted to test whether IN OXT and AVP were specific to peri-adolescent social behavior or if overnight social isolation increased their general anxiety. We tested the effect of IN OXT and IN AVP on two nonsocial tasks that assess anxiety and exploration: the elevated plus maze and the novel object task. Previous studies have found that OXT reduces anxiety in prairie voles, (Sobota et al., 2015; Ayers et al., 2011; Bales et al., 2004; Hale et al., 2021) and AVP increases anxiety in rats, prairie voles, and house mice (Bredewold and Veenema, 2018; Whylings et al., 2021; Simmons et al., 2017). These findings underscore the possibility that OXT and AVP could influence anxiety in California mice as well (Cid-Jofré et al., 2021). We viewed OXT effects on anxiety as exploratory because male California mice show a blunted stress response (review by Lonstein et al., 2015; Harris et al., 2011) while females exhibit oxytocin effects on anxiety (Duque-Wilckens & Trainor, 2022; Duque-Wilckens et al., 2018; Duque-Wilckens et al., 2020; Williams et al., 2020; Steinman et al., 2019) and no studies have examined OXT effects on stress or anxiety in juvenile or peri adolescent California mice. We therefore had no *a priori* predictions on the effect of OXT or AVP on anxiety.".

This study is the first to examine how OXT and AVP influence the maintenance of offspring-parent social preferences during peri-adolescence. This work adds to the literature on the mechanisms underlying social bonds within family units, such as pair bonds and maternal and paternal bonds to their offspring.

2. Methods

2.1. Animals

For our focal animals, we used 99 female and male *P. californicus* aged postnatal day (PND) 24–26 in litters 5–16 from 17 different breeding pairs. PND 24–26 is early weaning in California mice because they are typically weaned on PND 28–30. PND 24–26 likely represents the very early stages of peri-adolescence in California mice. When mice are housed together in same sex groups, coat molting and uterus-to-body weight and testes-to-body weight ratios do not show changes until PND 50, and final markers of puberty (vaginal opening and increased progesterone for females; preputial separation and increased testosterone for males) are not fully present until PND 80–90 (Wright et al., 2020, bioRxiv). This developmental time course is delayed compared to other small rodents used in social behavior research, such as house mice (*Mus musculus*) and prairie voles (*Microtus ochrogaster*). Both of these species are typically weaned around PND 21 (Bechard & Mason, 2010; Guoynes et al., 2018; Horii-Hayashi et al., 2013), and house mice also show changes in the vaginal opening (Drickamer, 1977) and penile maturation earlier (Purkart et al., 2020)). These animals were divided into two groups, with 44 animals in Group 1 that were randomly assigned to three treatment groups (OXT, AVP, and CTRL) and 55 animals in Group 2 that were randomly assigned to three treatment groups (OXT, AVP, and CTRL). For stimulus animals in the Group 1 study, we used 48 peri-adolescent stimulus animals aged PND 24–31 and 17 sets of parents (34 mice) aged 10–24 months that were unrelated by two generations. Parents were reused across the study, but never more than five times as

stimulus animals, and offspring from one set of parents were never assigned to the same treatment group. One to 11 pups were used from each breeder pair, with a mean of 5.94 and a standard deviation of 2.51 pups used per breeder pair. Before testing, peri-adolescent California mice were housed in breeding cages with their parents and 2–4 siblings (48 × 27 × 16 cm) under a 14L: 10D light cycle with lights off at 1:00 pm. For testing, peri-adolescent mice were randomly assigned to Group 1 and Group 2, with Group 1 exposed to a parent-peer preference test and Group 2 exposed to an elevated plus maze test and novel object test. Individual mice were then randomly assigned to the following treatments (described in detail below) with the stated sample sizes. In Group 1, sample sizes were: CTRL female (N = 8), OXT female (N = 8), AVP female (N = 7), CTRL male (N = 7), OXT male (N = 8), and AVP male (N = 6). In Group 2 sample sizes were: CTRL female (N = 10), OXT female (N = 10), AVP female (N = 9), CTRL male (N = 8), OXT male (N = 9), AVP male (N = 9). Animals were maintained per the National Institute of Health Guide for the Care and Use of Laboratory Animals and the University of Wisconsin-Madison Institutional Animal Care and Use Committee approved this research.

2.2. Intranasal oxytocin (OXT) and vasopressin (AVP) preparation

Female and male mice were infused intranasally with 17.5 uL of either sterile saline, OXT (0.5 IU/kg), or AVP (0.5 IU/kg) (Bachem, Torrance, California) (as used in Guoynes & Marler, 2021, 2022; Monari et al., 2021). The 0.5 IU/kg dose is relatively low and acts like an acute pulse or bolus release of neuropeptide (Guoynes & Marler, 2021; Monari et al., 2021), and is an order of magnitude lower than high doses and similar to the medium dose tested in a dose–response study in prairie voles (Bales et al., 2013: Guoynes et al., 2018). It is equivalent to the medium dose in an unpublished AVP dose–response study we conducted in adult California mice (Guoynes et al., 2023). This dose is also analogous to those used in human clinical trials (Anagnostou et al., 2014; Huang et al., 2021). OXT and AVP were dissolved in saline, prepared in two separate large batches, aliquoted into small plastic tubes, and frozen at 20 °C. Treatments, including saline control, were defrosted just before administration. A blunt cannula needle (33-gauge, 2.8 mm length; Plastics One, Roanoke, Virginia) was attached to cannula tubing, flushed, filled with the compound, then attached to an airtight Hamilton syringe (Bachem, Torrance, California). The animal was scruffed, and 17.5 uL of the compound was expelled dropwise through the cannula needle and allowed to absorb into the nasal mucosa (~10–20 s). One person conducted all IN administrations throughout the study to maintain consistency in handling and IN infusion. We chose to use IN delivery method because it has been shown to reach the brain in several species (Lee et al., 2020; Smith et al., 2019; Neumann et al., 2013; Striepens et al., 2013; Lee et al., 2018; Oppong-Damoah et al., 2019; Freeman et al., 2016; Quintana et al., 2021) and is less invasive than intracranial injections (see Guoynes & Marler, 2021; Guoynes & Marler, 2022).

2.3. Behavioral tests

To assess peri-adolescent social preference and exploration, we assigned peri-adolescent mice to one of two groups. Regardless of group assignment, each mouse was weaned from its home cage 24 h prior to testing (PND 23–25) and singly housed in a new home cage (30 × 19 × 13 cm). In Group 1, mice were tested in a 30-min parent-peer preference test as described below. In Group 2, mice were tested in the elevated plus maze test and then immediately tested in the novel object test for a cumulative total of 15 min of behavioral testing as described below (Fig. 1).

2.4. Parent-peer preference test

Each Group 1 mouse was tested in the parent-peer preference test using a three-chambered apparatus (91 cm × 46 cm × 43 cm) divided into three equal chambers. Each side chamber had a wire mesh partition at the back (30 cm × 10 cm) where stimulus animals could be presented.

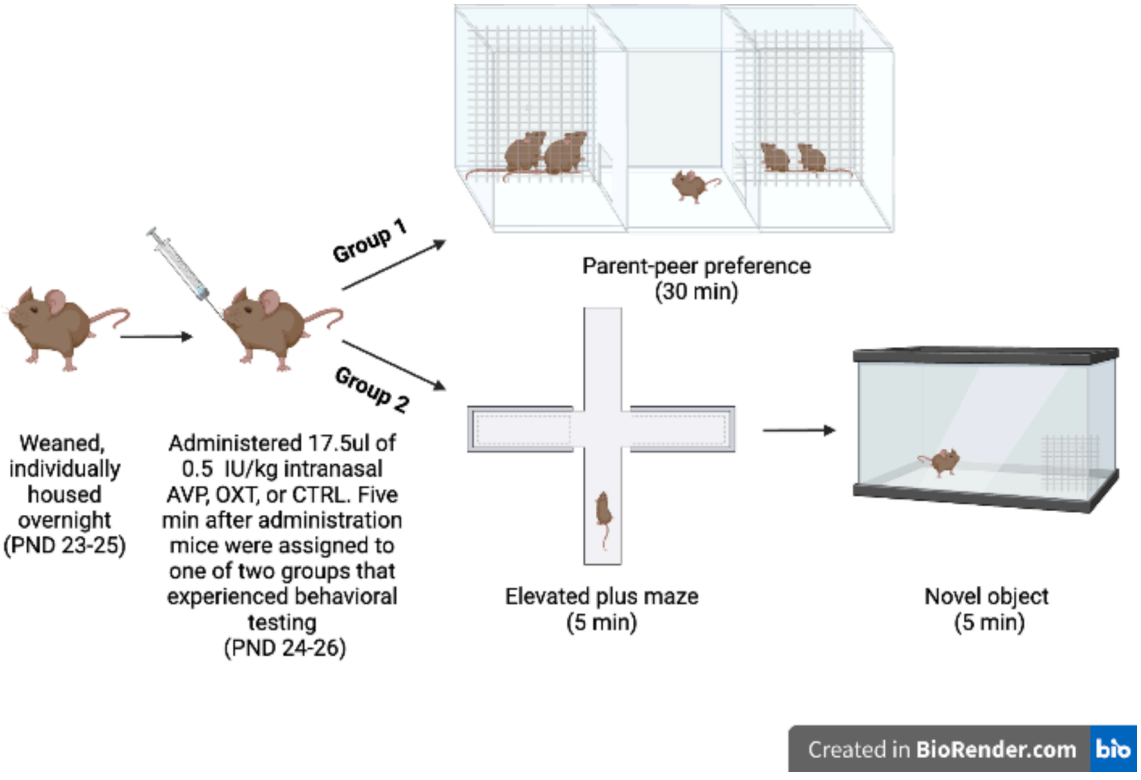

Fig. 1. Timeline and schematic for experimental design. All mice were weaned from their home cage, housed individually overnight, and randomly assigned an intranasal treatment. Approximately half of the mice were then assigned to Group 1, and the other half were assigned to Group 2.

Stimulus mice were as follows: the mother and father mice were placed on one side of the three-chambered Plexiglas cage behind the wire mesh, and a pair of age-matched female and male sibling mice (PND 24–31) to the focal mouse and its parents were placed on the other side of the apparatus. Similar tests have been previously used to assess social preferences in California mice in the Marler laboratory (Zhao & Marler, 2014; Zhao & Marler, 2016; Zhao et al., 2020) and in other labs to assess social preferences such as partner preference (Williams et al., 1992; Bales & Carter, 2003) and peer preference (Beery et al., 2018; Lee & Beery, 2021). Before each test, the parents of the focal mouse were randomly assigned to either the left or right side of the chamber, and unrelated, age-matched peers (one female, one male) were placed on the other side of the chamber. Once stimulus animals were placed in their respective chambers in the testing apparatus, each peri-adolescent mouse was given their randomly assigned IN dose (AVP, OXT, or CTRL) in their singly housed home cage. Five minutes after IN administration, the test mouse was placed in the center chamber of the testing apparatus, and its behavior was videotaped for 30 min. After this test, peri-adolescent mice were euthanized, and their brains were extracted for future studies.

2.5. Elevated plus maze test and novel object test

Mice assigned to Group 2 were first given the elevated plus maze test, followed by a novel object test. The maze consisted of two open and two enclosed opaque arms, each 67 cm long and 5.5 cm wide. The arms were elevated 1 m above the floor. peri-adolescent test mice were given IN treatment in their home cage five minutes prior to behavioral testing. At the start of the test, each mouse was placed in the center of the maze, and its behavior was videotaped for five min. Any animals that jumped off the open arms of the maze were captured and placed back in the center of the maze. Throughout the study, six mice fell off the apparatus, one time each. After a fall and placement back on the maze, mice explored the maze with a normal gait and spent time on both open and closed arms of the maze. Immediately following the elevated plus maze test, mice were moved to a new testing room and placed on the far side of a glass arena (50 cm × 30 cm × 30 cm) that contained a novel 5 cm × 5 cm × 5 cm metal cube, and behavior was recorded for five min. peri-adolescent mice were then euthanized, and their brains were extracted for future studies.

2.6. Behavior quantification

For an ethogram describing each behavior measured, see S. Table 1. For each behavior, we used a continuous sampling method. All experimenters were blind to treatment conditions during quantification. Behavior in all three tests was scored manually with a stopwatch.

2.7. Parent-peer preference test

For the trial to be successful, peri-adolescent mice had to visit both sides of the chamber in the first 10 min of the test to be scored. Two male mice did not meet this threshold and were excluded because they did not enter the chamber with the two peer mice. Time to enter each chamber and time spent in each of the three chambers were scored.

2.8. Elevated plus maze test

Trained observers scored behavior live during the test and recorded the duration of time in the open arms and the number of crosses through the center of the maze.

2.9. Novel object test

Behavior videos were scored for latency to approach the novel object, and time spent investigating the novel object was measured by the total number of seconds.

2.10. Data analysis

ANOVA tests were conducted for each behavioral test to compare saline control, OXT, and AVP treatment outcomes. The significance level was set at $p < 0.05$ for all analyses, and all tests were two-tailed. All reported p-values were corrected using Benjamini-Hochberg false discovery rate corrections to control for multiple comparisons when the effect of an X variable was tested for a relationship with multiple Y variables.

3. Results

3.1. Parent-peer preference test

We conducted a three-chambered social preference test to assess the effects of an acute pulse of OXT or AVP on juvenile social preference for their parents versus novel peers. Regardless of treatment, juvenile females preferred their parents over their peers ($F_{1,22} = 25.09$, $p < 0.0001$, $\Delta R^2 = 0.53$), and their peers over the empty middle chamber ($F_{1,22} = 55.38$, $p < 0.0001$, $\Delta R^2 = 0.72$) (Fig. 2**A**). Likewise, regardless of treatment, juvenile males preferred their parents over their peers ($F_{1,20} = 37.16$, $p < 0.0001$, $\Delta R^2 = 0.65$), and their peers over the empty chamber ($F_{1,20} = 25.15$, $p < 0.0001$, $\Delta R^2 = 0.55$) (Fig. 2**B**). To test for the effects of IN OXT and AVP on social preference, we created a preference score by subtracting the total percentage of each individual's preference for their peers from their preference for their parents [(time spent with parents/total test time)-(time spent with peers/total test time)]. There were no effects of treatment on the social preference expressed by females ($F_{2,\ 20} = 1.38$, $p = 0.27$) (Fig. 2**C**). However, IN OXT increased juvenile male preference for their parents ($F_{2,\ 18} = 4.98$, $p < 0.05$, $\Delta R^2 = 0.3562$) (Fig. 2**D**). There were no effects of treatment on preference for peers over an empty chamber expressed in females ($F_{2,\ 20} = 0.56$, $p = 0.58$) or males ($F_{2,\ 18} = 0.16$, $p = 0.85$) (Fig. 2**E-F**).

3.2. Elevated plus maze test

The goal was to assess the effects of OXT or AVP on juvenile exploration. There were no treatment effects of time spent on the open arms for females ($F_{2,26} = 0.31$, $p = 0.74$) or males ($F_{2,23} = 1.37$, $p = 0.27$) (Fig. 3**A**). There were also no treatment effects on the number of crosses through the center of the apparatus for females ($F_{2,26} = 0.20$, $p = 0.82$) or males ($F_{2,23} = 0.47$, $p = 0.63$) (Fig. 3**B**).

3.3. Novel object test

We measured behavior in a novel object task to assess the effects of an acute pulse of OXT or AVP on juvenile response to novelty. There were no treatment effects of latency to approach the novel object for females ($F_{2,16} = 0.94$, $p = 0.41$) or males ($F_{2,18} = 0.98$, $p = 0.40$) (Fig. 3**C**). There were also no treatment effects of time spent interacting with the novel object for females ($F_{2,16} = 0.01$, $p = 0.99$) or males ($F_{2,18} = 1.06$, $p = 0.37$) (Fig. 3**D**). There were also no differences in self-grooming for females ($F_{2,16} = 0.23$, $p = 0.79$) or males ($F_{2,18} = 1.56$, $p = 0.24$) (Fig. 3**E**). Finally, there were no differences in locomotor activity for females ($F_{2,16} = 0.12$, $p = 0.89$) or males ($F_{2,18} = 0.04$, $p = 0.96$) (Fig. 3**F**).

4. Discussion

Adolescence is a time of profound physiological and social change; however, the neural underpinnings of these social changes are just beginning to be explored (Pfeifer & Allen, 2021). To our knowledge, this is the first study to test the influence of a hormonal substrate on social preferences for parents versus peers in peri-adolescents. This study

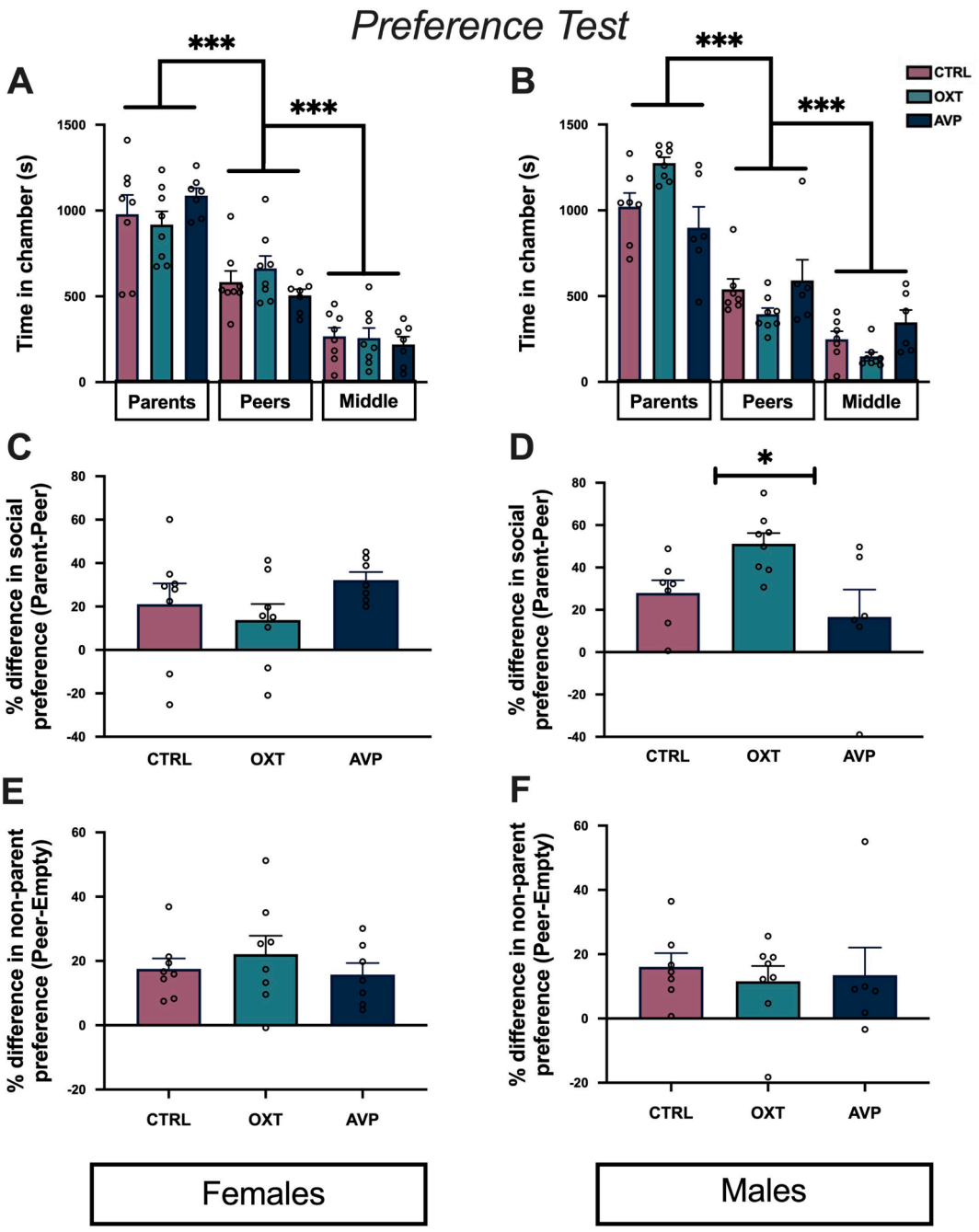

Fig. 2. (A) Both female and (B) male juveniles preferred their parents compared to their peers or the empty chamber and preferred their peers over an empty chamber. (C) Neither IN OXT nor IN AVP influenced social preference in female juveniles. (D) IN OXT increased juvenile male preference for their parents above and beyond their natural preference. (E) Neither IN OXT nor IN AVP inlfuenced preference for peers over an empty chamber in female juveniles. (F) Neither IN OXT nor IN AVP inlfuenced preference for peers over an empty chamber in male juveniles.

found that OXT but not AVP increased social preference in peri-adolescent males for their parents. Female peri-adolescents did not change their social preferences in response to either OXT or AVP, suggesting that mechanisms other than increases in exogenous OXT or AVP likely have greater impacts on female social preferences during peri-adolescence. It is also possible that endogenous levels of neuropeptides, differences in receptor densities, or reciprocal receptor binding are driving the differences between sexes we found in this study (Barberis & Tribollet, 1996; Alaerts et al., 2019; Wang et al., 1997). For example, this could include a ceiling threshold for the neuropeptides. Because neither OXT nor AVP influenced performance in the elevated plus maze task nor the novel object task, we suggest that the effect of OXT on males was not due to a decrease in general anxiety or sensation-seeking, which is also heightened during the peri-adolescent window in many species (Arnett, 1996; Stansfield et al., 2004). Future studies using agonists at different doses or OXT and AVP antagonists would be valuable to provide further insights into the mechanisms driving peri-adolescent social preference.

The results raise two issues which are: why is there a sex difference, and why is only OXT influencing behavior? While female and male California mice are very similar in their behaviors, one notable ecological difference occurs in behavior around the time of adolescence; females disperse farther from the natal territory than males (Ribble, 1992). In contrast to California mice, the males of most other

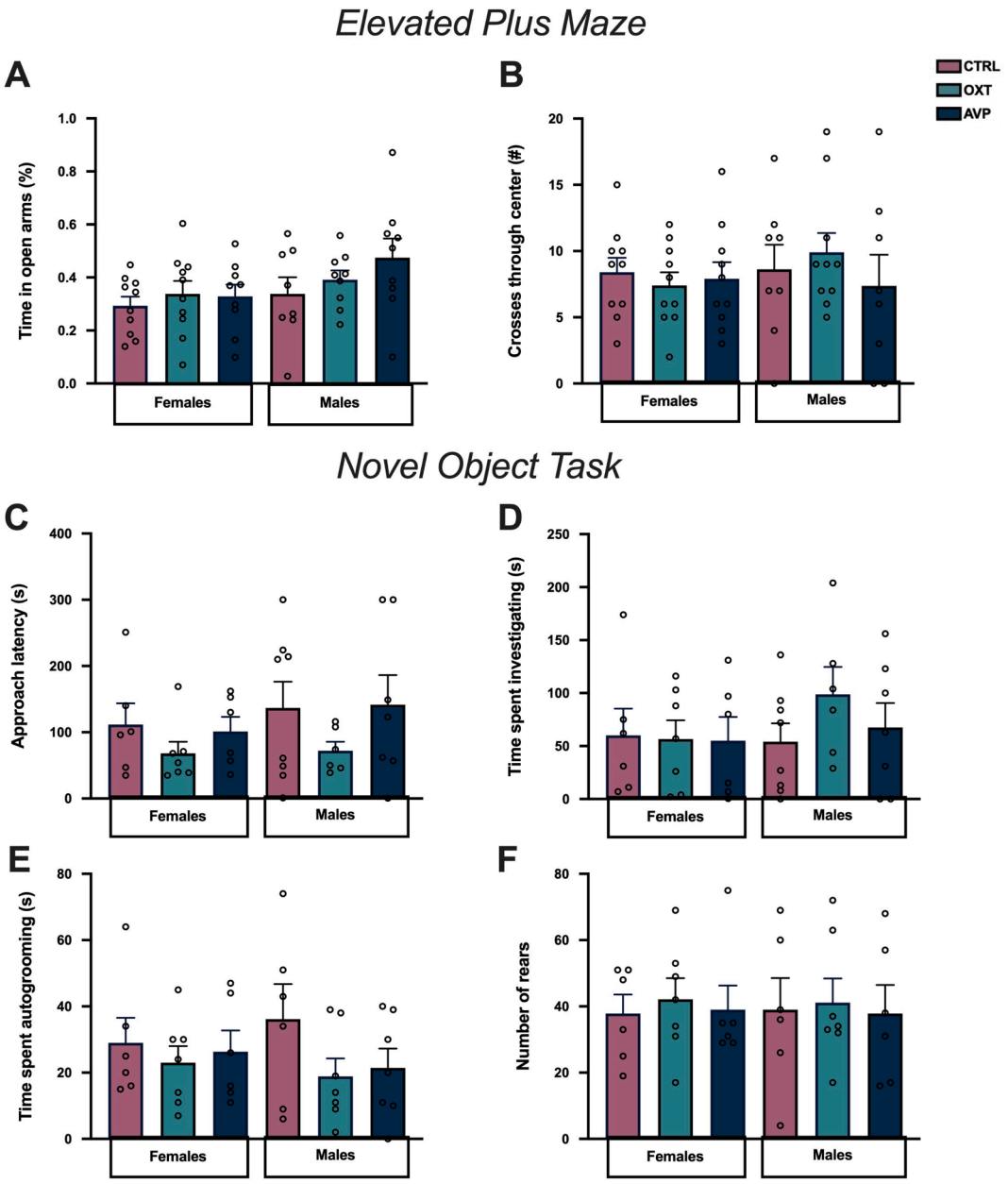

Fig. 3. (A) Neither IN OXT nor IN AVP influenced time spent on open arms (B) Neither IN OXT nor IN AVP influenced the number of crosses through the center of the elevated plus maze (C) Neither IN OXT nor AVP influenced latency to approach novel object (D) Neither IN OXT nor IN AVP influenced time spent investigating the novel object. (E) Neither IN OXT nor IN AVP influenced time spent autogrooming. (F) Neither IN OXT nor IN AVP influenced number of rears.

mammalian species are the ones that typically disperse farther from the natal territory (Pocock et al., 2005; Thompson, 2009). California mouse males can even take over their father's territory (personal communication, R. Petric). Anecdotal evidence in our lab suggests that when weaning a cage of juveniles at postnatal day 28, males are more likely to be still suckling on their mothers than females, possibly providing future insight into the sex differences we observed in this study. We speculate that males may require more plasticity in mechanisms influencing the parent-offspring bond, such as continued responsiveness to oxytocin to allow adjustments in social tolerance or bonding. While many species have sex differences, there is a general trend that females are more responsive to OXT manipulations, and males are more responsive to AVP manipulations such as in prairie voles (Liu and Wang, 2003; Lim & Young, 2004), there is a lack of comprehensive analysis of OXT and AVP system differences across the sexes in different model rodent species often used in labs that study social behavior (reviews by Dumais & Veenema, 2016; Lu et al., 2019; Lu & Hu, 2021). It is important to note that human males respond to both intranasal OXT and AVP (Hall et al., 2012; Rilling et al., 2014; Li et al., 2017; Guastella et al., 2010; Zhuang et al., 2021). Previous intranasal OXT studies in California mice have found that OXT influences male behavior (Guoynes & Marler, 2022; Steinman et al., 2016). Other rodent models, such as rats, house mice, and prairie voles, may have important differences in OXT and AVP receptor and ligand densities compared to California mice. Even relatively small differences in receptor expression are associated with sex differences and behavioral effects (Dumais & Veenema, 2016). We cannot exclude the possibility that the sex difference in response to OXT in these peri-adolescent mice is specific to California mice and future studies will be needed to further establish such findings.

Interestingly, we found that neither OXT nor AVP influenced performance on the elevated plus maze or in the novel object task, two tests that can measure stress and anxiety. There is evidence in many species

that OXT is generally anxiolytic and can suppress amygdala activity (Wahis et al., 2021; László et al., 2020) and that AVP is often anxiogenic and can increase amygdala activity (Hernández-Pérez et al., 2018; Harper et al., 2019). Amygdala activity in adolescents is often different than adults (Ferrara et al., 2021; Koss et al., 2014; Scherf et al., 2013; Juraska et al., 2013; Zimmermann et al., 2019), so it is possible that we did not see effects due to the age of the mice. Recent reviews also suggest that the relationship between OXT and AVP on anxiety may be more complex: social salience can lead to variability in both OXT and AVP's effects on stress and anxiety (MacDonald & Feifel, 2014; Jurek & Meyer, 2020). Our results suggest that that OXT or AVP do not influence general anxiety after overnight isolation for the dose used, but we cannot exclude the possibility that these neuropeptides influence social anxiety at this age.

However, an important limitation of this study is that we used an acute dose at a single time point. Chronic administration of OXT or AVP could lead to different behavioral results. Chronic administration at a similar dose used in this study in titi monkeys found that OXT treatment blunted male preference for their parents and increased time spent near unfamiliar pairs, and enhanced female preference for their parents (del Razo et al., 2020). Administering higher or lower doses of OXT may also have an impact on the behavioral response, as many studies have found contradictory effects of OXT and AVP at different points on the dose–response curve (Bales et al., 2013; Benelli et al., 1995; Martins et al., 2022; Borland et al., 2019; van Wimersma Greidanus and Maigret, 1996). However, similar acute doses of OXT have resulted in biologically relevant behavioral changes in other studies using adult California mice (Guoynes & Marler, 2022; Steinman et al., 2016; Monari et al., 2021). We also conducted a dose–response study with intranasal AVP in adult California mice and found that this 0.5 IU/kg dose increased aggression at pairing (Guoynes et al., 2023).

Regardless of the reason for why there were sex differences in the response of male and female juveniles to OXT, there are several speculative reasons why OXT made peri-adolescent males prefer to spend more time with their parents. One possibility is that IN OXT administration reinforces the parent-offspring social bond. In this study, the peri-adolescents were removed from their home cage for 24 h prior to testing; the loss of physical contact with their natal nest and parents could have caused OXT levels to decrease by removing the touch, smell, and gaze components of the social bond that reinforce the positive feedback release of OXT (Neumann et al., 1996; Romero et al., 2014; Wang et al., 2022), and IN OXT could have reinstated this positive feedback signaling. Studies in prairie voles have shown that adult reproductive and non-reproductive social behavior can be altered by the quality of the natal rearing, suggesting that the parent-offspring social interactions meaningfully impact the brain and behavior (Ahern et al., 2021; Perkeybile et al., 2019; Perkeybile & Bales, 2017; Bales et al., 2018). More broadly, for males, OXT and familiarity may be driving the heightened preference. OXT can increase the memory of and attention and affection towards familiar individuals (Marsh et al., 2021; Rimmele et al., 2009; Lu et al., 2019; Scheele et al., 2013). Therefore, we cannot rule out the possibility that familiarity is driving social preference for their parents. However, if the change in social preference is driven by familiarity, it is unlikely to be caused by anxiety because OXT did not influence elevated plus behavior. This is consistent with a study in prairie voles that found philopatry was not linked to high anxiety (del Razo & Bales, 2016). It is also unlikely that novelty avoidance led to the change in social preference because there was no difference in novel object task performance and the OXT male mice spent ~43 % more time in the peer chamber than in the empty chamber where they could avoid novel social contact. This increase is similar to control-treated males (~46 %). For the above reasons, we argue that OXT is enhancing prosocial behavior on the basis of the offspring-parent bond and/or familiarity.

Again, we can only speculate about the sex differences and why AVP is not influencing behavior during peri-adolescence. It is, however, interesting to note that adult male, but not female, California mice have greater OXT receptor binding in the cingulate cortex and bed nucleus of the stria terminalis, two areas known for emotion processing and social behavior. In contrast, there is no difference in AVP receptor binding (Insel et al., 1991). Sex differences in response to neuropeptides are often found, but it has been difficult to identify why this occurs. As the functions of neuropeptides during peri-adolescence are understudied, more research will be needed to examine functions, especially since neuropeptide effects on behavior can be context-dependent (see Reviews by Carter et al., 2020; Caldwell, 2018; Bartz et al., 2011; Rieger et al., 2022).

The neurobiological underpinnings of social bond maintenance in peri-adolescence are important to study because they offer a unique window into the factors associated with changing social preferences. Mammalian offspring are born and immediately initiate a social bond with their mother and possibly both parents in bi-parental species. However, the maintenance of these social bonds is tentative and depends on experience, environmental state, and biological substrate signaling. Most mammals, including humans, will start to lose their social preference for their parents over time. In peri-adolescent humans, activation of the nucleus accumbens to the ventromedial prefrontal cortex circuit is dependent on age: in younger adolescents (~7–12 years old), it is activated in response to their mother's voice, but in older adolescents (~13–16 years old) it is activated by the voices of strangers (Abrams et al., 2022). Because the nucleus accumbens to the ventromedial prefrontal cortex is known for both reward and social evaluation, these results underscore the importance of social sensory signals triggering the expression of social preferences.

This study is the first to demonstrate that OXT influences the maintenance of offspring-parent social preferences during peri-adolescence in males but not females. As such, this finding suggests that sex differences may also play a role in the mechanisms underlying social bond maintenance within family units. This work adds to the growing body of literature on social bonds within family units and with unrelated conspecifics.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ygcen.2023.114230.

References

Abrams, D.A., Mistry, P.K., Baker, A.E., Padmanabhan, A., Menon, V., 2022. A neurodevelopmental shift in reward circuitry from mother's to nonfamilial voices in adolescence. J. Neurosci. 42 (20), 4164–4173.

Acevedo-Rodriguez, A., Mani, S.K., Handa, R.J., 2015. Oxytocin and estrogen receptor β in the brain: an overview. Front. Endocrinol. 6, 160.

Ahern, T.H., Olsen, S., Tudino, R., Beery, A.K., 2021. Natural variation in the oxytocin receptor gene and rearing interact to influence reproductive and nonreproductive social behavior and receptor binding. Psychoneuroendocrinology 128, 105209.
Alaerts, K., Bernaerts, S., Vanaudenaerde, B., Daniels, N., Wenderoth, N., 2019. Amygdala–hippocampal connectivity is associated with endogenous levels of oxytocin and can be altered by exogenously administered oxytocin in adults with autism. Biol. Psychiatry: Cognitive Neurosci. Neuroimag. 4 (7), 655–663.
Albers, H.E., 2012. The regulation of social recognition, social communication and aggression: vasopressin in the social behavior neural network. Horm. Behav. 61 (3), 283–292.
Albers, H.E., Bamshad, M., 1999. Role of vasopressin and oxytocin in the control of social behavior in Syrian hamsters (Mesocricetus auratus. Prog. Brain Res. 119, 395–408.
Anagnostou, E., Soorya, L., Brian, J., Dupuis, A., Mankad, D., Smile, S., Jacob, S., 2014. Intranasal oxytocin in the treatment of autism spectrum disorders: a review of literature and early safety and efficacy data in youth. Brain Res. 1580, 188–198.
Arnett, J.J., 1996. Sensation seeking, aggressiveness, and adolescent reckless behavior. Pers. Individ. Differ. 20 (6), 693–702.
Ayers, L.W., Missig, G., Schulkin, J., Rosen, J.B., 2011. Oxytocin reduces background anxiety in a fear-potentiated startle paradigm: peripheral vs central administration. Neuropsychopharmacology 36 (12), 2488–2497.
Bales, K.L., Carter, C.S., 2003. Sex differences and developmental effects of oxytocin on aggression and social behavior in prairie voles (Microtus ochrogaster). Horm. Behav. 44 (3), 178–184.
Bales, K.L., Pfeifer, L.A., Carter, C.S., 2004. Sex differences and developmental effects of manipulations of oxytocin on alloparenting and anxiety in prairie voles. Developm. Psychobiol.: J. Int. Soc. Develop. Psychobiol. 44 (2), 123–131.
Bales, K.L., Witczak, L.R., Simmons, T.C., Savidge, L.E., Rothwell, E.S., Rogers, F.D., Manning, R.A., Heise, M.J., Englund, M., Del Razo, R.A., 2018. Social touch during development: Long-term effects on brain and behavior. Neurosci. Biobehav. Rev. 95, 202–219.
Barberis, C., Tribollet, E., 1996. Vasopressin and oxytocin receptors in the central nervous system. Criti. Rev.™ Neurobiology 10 (1).
Bartz, J.A., Zaki, J., Bolger, N., Ochsner, K.N., 2011. Social effects of oxytocin in humans: context and person matter. Trends Cogn. Sci. 15 (7), 301–309.
Baxter, A., Anderson, M., Seelke, A.M., Kinnally, E.L., Freeman, S.M., Bales, K.L., 2020. Oxytocin receptor binding in the titi monkey hippocampal formation is associated with parental status and partner affiliation. Sci. Rep. 10 (1), 1–14.
Bechard, A., Mason, G., 2010. Leaving home: a study of laboratory mouse pup independence. Appl. Anim. Behav. Sci. 125 (3–4), 181–188.
Beery, A.K., Christensen, J.D., Lee, N.S., Blandino, K.L., 2018. Specificity in sociality: mice and prairie voles exhibit different patterns of peer affiliation. Front. Behav. Neurosci. 12, 50.
Benelli, A., Bertolini, A., Poggioli, R., Menozzi, B., Basaglia, R., Arletti, R., 1995. Polymodal dose-response curve for oxytocin in the social recognition test. Neuropeptides 28 (4), 251–255.
Bester-Meredith, J.K., Marler, C.A., 2001. Vasopressin and aggression in cross-fostered California mice (Peromyscus californicus) and white-footed mice (Peromyscus leucopus). Horm. Behav. 40 (1), 51–64.
Bester-Meredith, J.K., Young, L.J., Marler, C.A., 1999. Species differences in paternal behavior and aggression in Peromyscus and their associations with vasopressin immunoreactivity and receptors. Horm. Behav. 36 (1), 25–38.
Borland, J.M., Rilling, J.K., Frantz, K.J., Albers, H.E., 2019. Sex-dependent regulation of social reward by oxytocin: an inverted U hypothesis. Neuropsychopharmacology 44 (1), 97–110.
Bredewold, R., Veenema, A.H., 2018. Sex differences in the regulation of social and anxiety-related behaviors: insights from vasopressin and oxytocin brain systems. Curr. Opin. Neurobiol. 49, 132–140.
Brenhouse, H.C., Andersen, S.L., 2011. Developmental trajectories during adolescence in males and females: a cross-species understanding of underlying brain changes. Neurosci. Biobehav. Rev. 35 (8), 1687–1703.
Caldwell, H.K., 2018. Oxytocin and sex differences in behavior. Curr. Opin. Behav. Sci. 23, 13–20.
Carter, C.S., Williams, J.R., Witt, D.M., Insel, T.R., 1992. Oxytocin and social bonding. Ann. N. Y. Acad. Sci. 652, 204–211.
Carter, C.S., Kenkel, W.M., MacLean, E.L., Wilson, S.R., Perkeybile, A.M., Yee, J.R., Ferris, C.F., Nazarloo, H.P., Porges, S.W., Davis, J.M., Connelly, J.J., 2020. Is oxytocin "nature's medicine". Pharmacol. Rev. 72 (4), 829–861.
Cid-Jofré, V., Moreno, M., Reyes-Parada, M., Renard, G.M., 2021. Role of oxytocin and vasopressin in neuropsychiatric disorders: therapeutic potential of agonists and antagonists. Int. J. Mol. Sci. 22 (21), 12077.
Crockford, C., Wittig, R.M., Langergraber, K., Ziegler, T.E., Zuberbühler, K., Deschner, T., 2013. Urinary oxytocin and social bonding in related and unrelated wild chimpanzees. Proc. R. Soc. B Biol. Sci. 280 (1755), 20122765.
Curley, J.P., Jensen, C.L., Franks, B., Champagne, F.A., 2012. Variation in maternal and anxiety-like behavior associated with discrete patterns of oxytocin and vasopressin 1a receptor density in the lateral septum. Horm. Behav. 61 (3), 454–461.
de Moura Oliveira, V.E., Neumann, I.D., de Jong, T.R., 2019. Post-weaning social isolation exacerbates aggression in both sexes and affects the vasopressin and oxytocin system in a sex-specific manner. Neuropharmacology 156, 107504.
De Vries, G.J., Wang, Z., Bullock, N.A., Numan, S., 1994. Sex differences in the effects of testosterone and its metabolites on vasopressin messenger RNA levels in the bed nucleus of the stria terminalis of rats. J. Neurosci. 14 (3), 1789–1794.
Del Razo, R.A., Bales, K.L., 2016. Exploration in a dispersal task: effects of early experience and correlation with other behaviors in prairie voles (Microtus ochrogaster). Behav. Process. 132, 66–75.
Del Razo, R.A., Berger, T., Conley, A.J., Freeman, S.M., Goetze, L.R., Jacob, S., Lawrence, R.H., Mendoza, S.P., Rothwell, E.S., Savidge, L.E., Solomon, M., 2020. Effects of chronic intranasal oxytocin on behavior and cerebral glucose uptake in juvenile titi monkeys. Psychoneuroendocrinology 113, 104494.
Delevich, K., Klinger, M., Okada, N.J., Wilbrecht, L., 2021. Coming of age in the frontal cortex: the role of puberty in cortical maturation. In: Seminars in Cell & Developmental Biology, Vol. 118. Academic Press, pp. 64–72.
Drickamer, L.C., 1977. Seasonal variation in litter size, bodyweight and sexual maturation in juvenile female house mice (Mus musculus). Lab. Anim 11 (3), 159–162.
Duell, N., Steinberg, L., 2019. Positive risk-taking in adolescence. Child Dev. Perspect. 13 (1), 48–52.
Dumais, K.M., Veenema, A.H., 2016. Vasopressin and oxytocin receptor systems in the brain: sex differences and sex-specific regulation of social behavior. Front. Neuroendocrinol. 40, 1–23.
Duque-Wilckens, N., Steinman, M.Q., Busnelli, M., Chini, B., Yokoyama, S., Pham, M., Laredo, S.A., Hao, R., Perkeybile, A.M., Minie, V.A., Tan, P.B., 2018. Oxytocin receptors in the anteromedial bed nucleus of the stria terminalis promote stress-induced social avoidance in female California mice. Biol. Psychiatry 83 (3), 203–213.
Duque-Wilckens, N., Trainor, B.C., 2022. Oxytocin release in stressful times. Psychoneuroendocrinology 140, 105709.
Duque-Wilckens, N., Torres, L.Y., Yokoyama, S., Minie, V.A., Tran, A.M., Petkova, S.P., Hao, R., Ramos-Maciel, S., Rios, R.A., Jackson, K., Flores-Ramirez, F.J., 2020. Extrahypothalamic oxytocin neurons drive stress-induced social vigilance and avoidance. Proc. Natl. Acad. Sci. 117 (42), 26406–26413.
Febo, M., Numan, M., Ferris, C.F., 2005. Functional magnetic resonance imaging shows oxytocin activates brain regions associated with mother–pup bonding during suckling. J. Neurosci. 25 (50), 11637–11644.
Ferrara, N.C., Trask, S., Avonts, B., Loh, M.K., Padival, M., Rosenkranz, J.A., 2021. Developmental shifts in amygdala activity during a high social drive state. J. Neurosci. 41 (45), 9308–9325.
Forbes, E.E., Dahl, R.E., 2010. Pubertal development and behavior: hormonal activation of social and motivational tendencies. Brain Cogn. 72 (1), 66–72.
Frayne, J., Nicholson, H.D., 1995. Effect of oxytocin on testosterone production by isolated rat Leydig cells is mediated via a specific oxytocin receptor. Biol. Reprod. 52 (6), 1268–1273.
Frazier, C.R., Trainor, B.C., Cravens, C.J., Whitney, T.K., Marler, C.A., 2006. Paternal behavior influences development of aggression and vasopressin expression in male California mouse offspring. Horm. Behav. 50 (5), 699–707.
Freeman, S.M., Samineni, S., Allen, P.C., Stockinger, D., Bales, K.L., Hwa, G.G., Roberts, J.A., 2016. Plasma and CSF oxytocin levels after intranasal and intravenous oxytocin in awake macaques. Psychoneuroendocrinology 66, 185–194.
Guastella, A.J., Kenyon, A.R., Alvares, G.A., Carson, D.S., Hickie, I.B., 2010. Intranasal arginine vasopressin enhances the encoding of happy and angry faces in humans. Biol. Psychiatry 67 (12), 1220–1222.
Gubernick, D.J., Winslow, J.T., Jensen, P., Jeanotte, L., Bowen, J., 1995. Oxytocin changes in males over the reproductive cycle in the monogamous, biparental California mouse, Peromyscus californicus. *Hormones Behavior* 29 (1), 59–73.
Guoynes, C.D., Corcoran, J., Kastar, E., Corradi, E., & Marler, C.A. (2023). Intranasal AVP increases aggression at courtship at low and medium doses in California mice (Peromyscus californicus).
Guoynes, C.D., Marler, C.A., 2021. An acute dose of intranasal oxytocin rapidly increases maternal communication and maintains maternal care in primiparous postpartum California mice. PLoS One 16 (4), e0244033.
Guoynes, C.D., Marler, C.A., 2022. Intranasal oxytocin reduces pre-courtship aggression and increases paternal response in California mice (Peromyscus californicus). Physiol. Behav. 249, 113773.
Guoynes, C.D., Simmons, T.C., Downing, G.M., Jacob, S., Solomon, M., Bales, K.L., 2018. Chronic intranasal oxytocin has dose-dependent effects on central oxytocin and vasopressin systems in prairie voles (Microtus ochrogaster). Neuroscience 369, 292–302.
Hale, L.H., Tickerhoof, M.C., Smith, A.S., 2021. Chronic intranasal oxytocin reverses stress-induced social avoidance in female prairie voles. Neuropharmacology 198, 108770.
Hall, S.S., Lightbody, A.A., McCarthy, B.E., Parker, K.J., Reiss, A.L., 2012. Effects of intranasal oxytocin on social anxiety in males with fragile X syndrome. Psychoneuroendocrinology 37 (4), 509–518.
Harper, K.M., Knapp, D.J., Butler, R.K., Cook, C.A., Criswell, H.E., Stuber, G.D., Breese, G.R., 2019. Amygdala arginine vasopressin modulates chronic ethanol withdrawal anxiety-like behavior in the social interaction task. Alcohol. Clin. Exp. Res. 43 (10), 2134–2143.
Harris, B.N., Perea-Rodriguez, J.P., Saltzman, W., 2011. Acute effects of corticosterone injection on paternal behavior in California mouse (Peromyscus californicus) fathers. Horm. Behav. 60 (5), 666–675.
Heinrichs, M., von Dawans, B., Domes, G., 2009. Oxytocin, vasopressin, and human social behavior. Front. Neuroendocrinol. 30 (4), 548–557.
Hernández-Pérez, O.R., Crespo-Ramírez, M., Cuza-Ferrer, Y., Anias-Calderón, J., Zhang, L., Roldan-Roldan, G., Aguilar-Roblero, R., Borroto-Escuela, D.O., Fuxe, K., Perez de la Mora, M., 2018. Differential activation of arginine-vasopressin receptor subtypes in the amygdaloid modulation of anxiety in the rat by arginine-vasopressin. Psychopharmacology (Berl) 235 (4), 1015–1027.
Horii-Hayashi, N., Sasagawa, T., Matsunaga, W., Matsusue, Y., Azuma, C., Nishi, M., 2013. Developmental changes in desensitisation of c-Fos expression induced by repeated maternal separation in pre-weaned mice. J. Neuroendocrinol. 25 (2), 158–167.

Huang, Y., Huang, X., Ebstein, R.P., Yu, R., 2021. Intranasal oxytocin in the treatment of autism spectrum disorders: A multilevel meta-analysis. Neurosci. Biobehav. Rev. 122, 18–27.
Insel, T.R., 1990. Regional changes in brain oxytocin receptors post-partum: time-course and relationship to maternal behaviour. J. Neuroendocrinol. 2 (4), 539–545.
Insel, T.R., Gelhard, R., Shapiro, L.E., 1991. The comparative distribution of forebrain receptors for neurohypophyseal peptides in monogamous and polygamous mice. Neuroscience 43 (2–3), 623–630.
Jarcho, M.R., Mendoza, S.P., Mason, W.A., Yang, X., Bales, K.L., 2011. Intranasal vasopressin affects pair bonding and peripheral gene expression in male *Callicebus cupreus*. Genes Brain Behav. 10 (3), 375–383.
Juraska, J.M., Sisk, C.L., DonCarlos, L.L., 2013. Sexual differentiation of the adolescent rodent brain: hormonal influences and developmental mechanisms. Horm. Behav. 64 (2), 203–210.
Jurek, B., Meyer, M., 2020. Anxiolytic and anxiogenic? How the transcription factor MEF2 might explain the manifold behavioral effects of oxytocin. Front. Endocrinol. 11, 186.
Keebaugh, A.C., Barrett, C.E., LaPrairie, J.L., Jenkins, J.J., Young, L.J., 2015. RNAi knockdown of oxytocin receptor in the nucleus accumbens inhibits social attachment and parental care in monogamous female prairie voles. Soc. Neurosci. 10 (5), 561–570.
Kendrick, K.M., 2000. Oxytocin, motherhood and bonding. *Experim. Physiol.*, *85*(s1), 111s-124s.
Kent, K., Arientyl, V., Khachatryan, M.M., Wood, R.I., 2013. Oxytocin induces a conditioned social preference in female mice. J. Neuroendocrinol. 25 (9), 803–810.
Keverne, E.B., Curley, J.P., 2004. Vasopressin, oxytocin and social behaviour. Curr. Opin. Neurobiol. 14 (6), 777–783.
Keverne, E.B., Kendrick, K.M., 1992. Oxytocin facilitation of maternal behavior in sheep. Ann. N. Y. Acad. Sci. 652 (1), 83–101.
Koolschijn, P.C.M., Peper, J.S., Crone, E.A., 2014. The influence of sex steroids on structural brain maturation in adolescence. PLoS One 9 (1), e83929.
Koss, W.A., Belden, C.E., Hristov, A.D., Juraska, J.M., 2014. Dendritic remodeling in the adolescent medial prefrontal cortex and the basolateral amygdala of male and female rats. Synapse 68 (2), 61–72.
László, K., Péczely, L., Géczi, F., Kovács, A., Zagoracz, O., Ollmann, T., Kertes, E., Kállai, V., László, B., Berta, B., Karádi, Z., 2020. The role of D2 dopamine receptors in oxytocin-induced place preference and anxiolytic effect. Horm. Behav. 124, 104777.
Lee, N.S., Beery, A.K., 2021. The role of dopamine signaling in prairie vole peer relationships. Horm. Behav. 127, 104876.
Lee, M.R., Scheidweiler, K.B., Diao, X.X., Akhlaghi, F., Cummins, A., Huestis, M.A., Leggio, L., Averbeck, B.B., 2018. Oxytocin by intranasal and intravenous routes reaches the cerebrospinal fluid in rhesus macaques: determination using a novel oxytocin assay. Mol. Psychiatry 23 (1), 115–122.
Lee, M.R., Shnitko, T.A., Blue, S.W., Kaucher, A.V., Winchell, A.J., Erikson, D.W., Grant, K.A., Leggio, L., 2020. Labeled oxytocin administered via the intranasal route reaches the brain in rhesus macaques. Nat. Commun. 11 (1), 1–10.
Li, T., Chen, X., Mascaro, J., Haroon, E., Rilling, J.K., 2017. Intranasal oxytocin, but not vasopressin, augments neural responses to toddlers in human fathers. Horm. Behav. 93, 193–202.
Lim, M.M., Young, L.J., 2004. Vasopressin-dependent neural circuits underlying pair bond formation in the monogamous prairie vole. Neuroscience 125 (1), 35–45.
Liu, Y., Wang, Z.X., 2003. Nucleus accumbens oxytocin and dopamine interact to regulate pair bond formation in female prairie voles. Neuroscience 121 (3), 537–544.
Lonstein, J.S., Pereira, M., Morrell, J.I., Marler, C.A., 2015. Parenting Behavior. Physiology of Reproduction, New York, pp. 2371–2437.
Loth, M.K., Donaldson, Z.R., 2021. Oxytocin, dopamine, and opioid interactions underlying pair bonding: highlighting a potential role for microglia. Endocrinology 162 (2), bqaa223.
Lu, Q., Hu, S., 2021. Sex differences of oxytocin and vasopressin in social behaviors. Handb. Clin. Neurol. 180, 65–88.
Lu, Q., Lai, J., Du, Y., Huang, T., Prukpitikul, P., Xu, Y., Hu, S., 2019. Sexual dimorphism of oxytocin and vasopressin in social cognition and behavior. Psychol. Res. Behav. Manag. 12, 337.
Lukas, M., Neumann, I.D., 2014. Social preference and maternal defeat-induced social avoidance in virgin female rats: sex differences in involvement of brain oxytocin and vasopressin. J. Neurosci. Methods 234, 101–107.
Lukas, M., Toth, I., Reber, S.O., Slattery, D.A., Veenema, A.H., Neumann, I.D., 2011. The neuropeptide oxytocin facilitates pro-social behavior and prevents social avoidance in rats and mice. Neuropsychopharmacology 36 (11), 2159–2168.
MacDonald, K., Feifel, D., 2014. Oxytocin′ s role in anxiety: A critical appraisal. Brain Res. 1580, 22–56.
Marsh, N., Scheele, D., Postin, D., Onken, M., Hurlemann, R., 2021. Eye-Tracking reveals a role of oxytocin in attention allocation towards familiar faces. Front. Endocrinol. 12, 629760.
Martins, D., Brodmann, K., Veronese, M., Dipasquale, O., Mazibuko, N., Schuschnig, U., Zelaya, F., Fotopoulou, A., Paloyelis, Y., 2022. "Less is more": A dose-response account of intranasal oxytocin pharmacodynamics in the human brain. Prog. Neurobiol. 211, 102239.
Maud, C., Ryan, J., McIntosh, J.E., Olsson, C.A., 2018. The role of oxytocin receptor gene (OXTR) DNA methylation (DNAm) in human social and emotional functioning: a systematic narrative review. BMC Psychiatry 18 (1), 1–13.
McCarthy, M.M., McDonald, C.H., Brooks, P.J., Goldman, D., 1996. An anxiolytic action of oxytocin is enhanced by estrogen in the mouse. Physiol. Behav. 60 (5), 1209–1215.
Monari, P.K., Rieger, N.S., Schefelker, J., Marler, C.A., 2021. Intranasal oxytocin drives coordinated social approach. Sci. Rep. 11 (1), 1–13.
Murakami, G., Hunter, R.G., Fontaine, C., Ribeiro, A., Pfaff, D., 2011. Relationships among estrogen receptor, oxytocin and vasopressin gene expression and social interaction in male mice. Eur. J. Neurosci. 34 (3), 469–477.
Nagasawa, M., Okabe, S., Mogi, K., Kikusui, T., 2012. Oxytocin and mutual communication in mother-infant bonding. Front. Hum. Neurosci. 6, 31.
Neumann, I., Ludwig, M., Engelmann, M., Pittman, Q.J., Landgraf, R., 1993. Simultaneous microdialysis in blood and brain: oxytocin and vasopressin release in response to central and peripheral osmotic stimulation and suckling in the rat. Neuroendocrinology 58 (6), 637–645.
Neumann, I., Douglas, A.J., Pittman, Q.J., Russell, J.A., Landgraf, R., 1996. Oxytocin released within the supraoptic nucleus of the rat brain by positive feedback action is involved in parturition-related events. J. Neuroendocrinol. 8 (3), 227–233.
Neumann, I.D., Maloumby, R., Beiderbeck, D.I., Lukas, M., Landgraf, R., 2013. Increased brain and plasma oxytocin after nasal and peripheral administration in rats and mice. Psychoneuroendocrinology 38 (10), 1985–1993.
Nishimori, K., Young, L.J., Guo, Q., Wang, Z., Insel, T.R., Matzuk, M.M., 1996. Oxytocin is required for nursing but is not essential for parturition or reproductive behavior. Proc. Natl. Acad. Sci. 93 (21), 11699–11704.
Oppong-Damoah, A., Zaman, R.U., D'Souza, M.J., Murnane, K.S., 2019. Nanoparticle encapsulation increases the brain penetrance and duration of action of intranasal oxytocin. Horm. Behav. 108, 20–29.
Pedersen, C.A., Caldwell, J.D., Walker, C., Ayers, G., Mason, G.A., 1994. Oxytocin activates the postpartum onset of rat maternal behavior in the ventral tegmental and medial preoptic areas. Behav. Neurosci. 108 (6), 1163.
Peper, J.S., Pol, H.H., Crone, E.A., Van Honk, J., 2011. Sex steroids and brain structure in pubertal boys and girls: a mini-review of neuroimaging studies. Neuroscience 191, 28–37.
Perea-Rodriguez, J.P., Takahashi, E.Y., Amador, T.M., Hao, R.C., Saltzman, W., Trainor, B.C., 2015. Effects of reproductive experience on central expression of progesterone, oestrogen α, oxytocin and vasopressin receptor mRNA in male California mice (Peromyscus californicus). J. Neuroendocrinol. 27 (4), 245–252.
Perkeybile, A.M., Bales, K.L., 2017. Intergenerational transmission of sociality: the role of parents in shaping social behavior in monogamous and non-monogamous species. J. Exp. Biol. 220 (1), 114–123.
Perkeybile, A.M., Carter, C.S., Wroblewski, K.L., Puglia, M.H., Kenkel, W.M., Lillard, T.S., Karaoli, T., Gregory, S.G., Mohammadi, N., Epstein, L., Bales, K.L., 2019. Early nurture epigenetically tunes the oxytocin receptor. Psychoneuroendocrinology 99, 128–136.
Pfeifer, J.H., Allen, N.B., 2021. Puberty initiates cascading relationships between neurodevelopmental, social, and internalizing processes across adolescence. Biol. Psychiatry 89 (2), 99–108.
Pocock, M.J., Hauffe, H.C., Searle, J.B., 2005. Dispersal in house mice. Biol. J. Linn. Soc. 84 (3), 565–583.
Prounis, G.S., Thomas, K., Ophir, A.G., 2018. Developmental trajectories and influences of environmental complexity on oxytocin receptor and vasopressin 1A receptor expression in male and female prairie voles. J. Comp. Neurol. 526 (11), 1820–1842.
Prugh, L.R., Deguines, N., Grinath, J.B., Suding, K.N., Bean, W.T., Stafford, R., Brashares, J.S., 2018. Ecological winners and losers of extreme drought in California. Nat. Clim. Chang. 8 (9), 819–824.
Purkart, L., Sigl-Glöckner, J., Brecht, M., 2020. Constant innervation despite pubertal growth of the mouse penis. J Comp Neurol 528 (13), 2269–2279.
Quintana, D.S., Lischke, A., Grace, S., Scheele, D., Ma, Y., Becker, B., 2021. Advances in the field of intranasal oxytocin research: lessons learned and future directions for clinical research. Mol. Psychiatry 26 (1), 80–91.
Ribble, D.O., 1992. Dispersal in a monogamous rodent, Peromyscus californicus. *Ecology* 73 (3), 859–866.
Rieger, N.S., Guoynes, C.D., Monari, P.K., Hammond, E.R., Malone, C.L., Marler, C.A., 2022. Neuroendocrine mechanisms of aggression in rodents. Motiv. Sci. 8 (2).
Rilling, J.K., DeMarco, A.C., Hackett, P.D., Chen, X., Gautam, P., Stair, S., Haroon, E., Thompson, R., Ditzen, B., Patel, R., Pagnoni, G., 2014. Sex differences in the neural and behavioral response to intranasal oxytocin and vasopressin during human social interaction. Psychoneuroendocrinology 39, 237–248.
Rimmele, U., Hediger, K., Heinrichs, M., Klaver, P., 2009. Oxytocin makes a face in memory familiar. J. Neurosci. 29 (1), 38–42.
Romeo, R.D., 2003. Puberty: a period of both organizational and activational effects of steroid hormones on neurobehavioural development. J. Neuroendocrinol. 15 (12), 1185–1192.
Romero, T., Nagasawa, M., Mogi, K., Hasegawa, T., Kikusui, T., 2014. Oxytocin promotes social bonding in dogs. Proc. Natl. Acad. Sci. 111 (25), 9085–9090.
Scheele, D., Wille, A., Kendrick, K.M., Stoffel-Wagner, B., Becker, B., Güntürkün, O., Maier, W., Hurlemann, R., 2013. Oxytocin enhances brain reward system responses in men viewing the face of their female partner. Proc. Natl. Acad. Sci. 110 (50), 20308–20313.
Scherf, K.S., Smyth, J.M., Delgado, M.R., 2013. The amygdala: an agent of change in adolescent neural networks. Horm. Behav. 64 (2), 298–313.
Shapiro, R.A., Xu, C., Dorsa, D.M., 2000. Differential transcriptional regulation of rat vasopressin gene expression by estrogen receptor α and β. Endocrinology 141 (11), 4056–4064.
Simmons, T.C., Balland, J.F., Dhauna, J., Yang, S.Y., Traina, J.L., Vazquez, J., Bales, K.L., 2017. Early intranasal vasopressin administration impairs partner preference in adult male prairie voles (Microtus ochrogaster). Front. Endocrinol. 8, 145.
Skuse, D.H., Lori, A., Cubells, J.F., Lee, I., Conneely, K.N., Puura, K., Lehtimäki, T., Binder, E.B., Young, L.J., 2014. Common polymorphism in the oxytocin receptor

gene (OXTR) is associated with human social recognition skills. Proc. Natl. Acad. Sci. 111 (5), 1987–1992.
Smith, A.S., Korgan, A.C., Young, W.S., 2019. Oxytocin delivered nasally or intraperitoneally reaches the brain and plasma of normal and oxytocin knockout mice. Pharmacol. Res. 146, 104324.
Smith, C.J., Poehlmann, M.L., Li, S., Ratnaseelan, A.M., Bredewold, R., Veenema, A.H., 2017. Age and sex differences in oxytocin and vasopressin V1a receptor binding densities in the rat brain: focus on the social decision-making network. Brain Struct. Funct. 222 (2), 981–1006.
Sobota, R., Mihara, T., Forrest, A., Featherstone, R.E., Siegel, S.J., 2015. Oxytocin reduces amygdala activity, increases social interactions, and reduces anxiety-like behavior irrespective of NMDAR antagonism. Behav. Neurosci. 129 (4), 389.
Spear, L. (2007). The Developing Brain and Adolescent-Typical Behavior Patterns An Evolutionary Approach. *Adolescent Psychopathology and the Developing Brain: Integrating Brain and Prevention Science*, 9.
Staes, N., Koski, S.E., Helsen, P., Fransen, E., Eens, M., Stevens, J.M., 2015. Chimpanzee sociability is associated with vasopressin (Avpr1a) but not oxytocin receptor gene (OXTR) variation. Horm. Behav. 75, 84–90.
Stansfield, K.H., Philpot, R.M., Kirstein, C.L., 2004. An animal model of sensation seeking: the adolescent rat. Ann. N. Y. Acad. Sci. 1021 (1), 453–458.
Steinberg, L., 2010. A dual systems model of adolescent risk-taking. Developm. Psychobiol. 52 (3), 216–224.
Steinman, M.Q., Duque-Wilckens, N., Greenberg, G.D., Hao, R., Campi, K.L., Laredo, S.A., Laman-Maharg, A., Manning, C.E., Doig, I.E., Lopez, E.M., Walch, K., 2016. Sex-specific effects of stress on oxytocin neurons correspond with responses to intranasal oxytocin. Biol. Psychiatry 80 (5), 406–414.
Steinman, M.Q., Duque-Wilckens, N., Trainor, B.C., 2019. Complementary neural circuits for divergent effects of oxytocin: social approach versus social anxiety. Biol. Psychiatry 85 (10), 792–801.
Striepens, N., Kendrick, K.M., Hanking, V., Landgraf, R., Wüllner, U., Maier, W., Hurlemann, R., 2013. Elevated cerebrospinal fluid and blood concentrations of oxytocin following its intranasal administration in humans. Sci. Rep. 3 (1), 1–5.
Taylor, J.H., Cavanaugh, J., French, J.A., 2017. Neonatal oxytocin and vasopressin manipulation alter social behavior during the juvenile period in Mongolian gerbils. Dev. Psychobiol. 59 (5), 653–657.
Taylor, J.H., Walton, J.C., McCann, K.E., Norvelle, A., Liu, Q., Vander Velden, J.W., Borland, J.M., Hart, M., Jin, C., Huhman, K.L. and Cox, D.N., 2022. CRISPR-Cas9 editing of the arginine–vasopressin V1a receptor produces paradoxical changes in social behavior in Syrian hamsters. *Proc. Natl. Acad. Sci., 119*(19), p.e2121037119.
Taylor, J.H., French, J.A., 2015. Oxytocin and vasopressin enhance responsiveness to infant stimuli in adult marmosets. Horm. Behav. 75, 154–159.
Thompson, E., 2009. The context of female dispersal in Kanyawara chimpanzees. Behaviour 146 (4–5), 629–656.
Trova, S., Bovetti, S., Bonzano, S., De Marchis, S., Peretto, P., 2021. Sex steroids and the shaping of the peripubertal brain: the sexual-dimorphic set-up of adult neurogenesis. Int. J. Mol. Sci. 22 (15), 7984.
van Wimersma Greidanus, T.B., Maigret, C., 1996. The role of limbic vasopressin and oxytocin in social recognition. Brain Res. 713 (1–2), 153–159.
Wahis, J., Baudon, A., Althammer, F., Kerspern, D., Goyon, S., Hagiwara, D., Lefevre, A., Barteczko, L., Boury-Jamot, B., Bellanger, B., Abatis, M., 2021. Astrocytes mediate the effect of oxytocin in the central amygdala on neuronal activity and affective states in rodents. Nat. Neurosci. 24 (4), 529–541.
Wang, F., Yin, X.S., Lu, J., Cen, C., Wang, Y., 2022. Phosphorylation-dependent positive feedback on the oxytocin receptor through the kinase PKD1 contributes to long-term social memory. Sci. Signal. 15 (719), eabd0033.
Wang, Z., Young, L.J., Liu, Y., Insel, T.R., 1997. Species differences in vasopressin receptor binding are evident early in development: comparative anatomic studies in prairie and montane voles. J Comp Neurol 378 (4), 535–546.
Wang, Z., Young, L.J., Insel, T.R., 1999. Voles and vasopressin: a review of molecular, cellular, and behavioral studies of pair bonding and paternal behaviors. Prog. Brain Res. 119, 483–499.
Whylings, J., Rigney, N., de Vries, G.J., Petrulis, A., 2021. Reduction in vasopressin cells in the suprachiasmatic nucleus in mice increases anxiety and alters fluid intake. Horm. Behav. 133, 104997.
Williams, J.R., Catania, K.C., Carter, C.S., 1992. Development of partner preferences in female prairie voles (Microtus ochrogaster): the role of social and sexual experience. Horm. Behav. 26 (3), 339–349.
Williams, A.V., Duque-Wilckens, N., Ramos-Maciel, S., Campi, K.L., Bhela, S.K., Xu, C.K., Jackson, K., Chini, B., Pesavento, P.A., Trainor, B.C., 2020. Social approach and social vigilance are differentially regulated by oxytocin receptors in the nucleus accumbens. Neuropsychopharmacology 45 (9), 1423–1430.
Winslow, J.T., Hastings, N., Carter, C.S., Harbaugh, C.R., Insel, T.R., 1993. A role for central vasopressin in pair bonding in monogamous prairie voles. Nature 365 (6446), 545–548.
Wood, R.I., Knoll, A.T., Levitt, P., 2015. Social housing conditions and oxytocin and vasopressin receptors contribute to ethanol-conditioned social preference in female mice. Physiol. Behav. 151, 469–477.
Wright, E.C., Culkin, H.I., Sekar, S., Kapoor, A., Corbett, C., & Trainor, B.C. (2020). Pubertal androgens reduce the effects of social stress on anxiety-related behaviors in California mice. *bioRxiv.*.
Yohn, C.N., Leithead, A.B., Becker, E.A., 2017. Increased vasopressin expression in the BNST accompanies paternally induced territoriality in male and female California mouse offspring. Horm. Behav. 93, 9–17.
Yohn, C.N., Leithead, A.B., Ford, J., Gill, A., Becker, E.A., 2018. Paternal care impacts oxytocin expression in California mouse offspring and basal testosterone in female, but not male pups. Front. Behav. Neurosci. 12, 181.
Young, L.J., Winslow, J.T., Wang, Z., Gingrich, B., Guo, Q., Matzuk, M.M., Insel, T.R., 1997. Gene targeting approaches to neuroendocrinology: oxytocin, maternal behavior, and affiliation. Horm. Behav. 31 (3), 221–231.
Young, L.J., Wang, Z., Donaldson, R., Rissman, E.F., 1998. Estrogen receptor α is essential for induction of oxytocin receptor by estrogen. Neuroreport 9 (5), 933–936.
Zhao, X., Castelli, F.R., Wang, R., Auger, A.P., Marler, C.A., 2020. Testosterone-related behavioral and neural mechanisms associated with location preferences: A model for territorial establishment. Horm. Behav. 121, 104709.
Zhao, X., Marler, C.A., 2014. Pair bonding prevents reinforcing effects of testosterone in male California mice in an unfamiliar environment. Proc. R. Soc. B Biol. Sci. 281 (1788), 20140985.
Zhao, X., Marler, C.A., 2016. Social and physical environments as a source of individual variation in the rewarding effects of testosterone in male California mice (Peromyscus californicus). Horm. Behav. 85, 30–35.
Zhuang, Q., Zheng, X., Becker, B., Lei, W., Xu, X., Kendrick, K.M., 2021. Intranasal vasopressin like oxytocin increases social attention by influencing top-down control, but additionally enhances bottom-up control. Psychoneuroendocrinology 133, 105412.
Zimmermann, K.S., Richardson, R., Baker, K.D., 2019. Maturational changes in prefrontal and amygdala circuits in adolescence: implications for understanding fear inhibition during a vulnerable period of development. Brain Sci. 9 (3), 65.