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Placentophagia in Weanling Female Laboratory Rats

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Abstract

Placentophagia is common in parturient mammals and offers physiological and behavioral advantages for mothers. In natural environments, weanlings are often present during the birth of younger siblings, but it is unknown if weanling rats are placentophagic or prefer placenta over other substances. To examine this, primiparous rats were remated during the postpartum estrus and weanling females remained in the nest during their mother's next parturition. Continuous observation revealed that 58% of weanlings were placentophagic. To determine if this placentophagia occurs away from parturient mothers, weanling females still living in their natal nest were offered placenta, liver, or cake frosting in a novel chamber. They ingested more placenta and liver than frosting. Thus, many weanling female laboratory rats are placentophagic during birth of younger siblings but do not selectively prefer placenta when tested outside their natal nest. Consequences of placentophagia by weanlings are unknown, but it may promote their alloparenting or postpartum mothering.

Keywords

alloparental; development; female; juvenile; maternal behavior; midwifery; placenta; puberty; rat

Placentophagia, the ingestion of placenta or fetal membranes and fluids, is commonly exhibited by most female mammals while they give birth (Kristal, DiPirro, & Thompson, 2012). Because placentophagia is so broadly displayed it is often suggested that it provides benefits to mothers and infants, which may include replenishing nutrients mothers lost during parturition, enhancing maternal attraction to newborns that promotes their earliest physical contact, potentiating the opioid signaling underlying peripartum analgesia and the emergence of caregiving behaviors, assisting neonatal breathing by removing suffocating membranes, and perhaps diminishing predator attraction to the bloodied nest site (Kristal, DiPirro, & Thompson, 2012). While female mammals are tremendously attracted to placenta and amniotic fluid immediately before, during, and after parturition, many or most nulliparous animals are thought to find afterbirth repugnant (Engwall & Kristal, 1977; Kristal & Graber, 1976; Kristal & Williams, 1973; Levy, Poindroin & Le Neindre 1983; Melo & Gonzalez-Mariscal, 2003).

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Studies by Wynne-Edwards and colleagues, however, have demonstrated that parturient females are not necessarily the only avid participants in placentophagia. In the socially monogamous and biparental Djungarian hamster, *Phodopus campbelli*, sires readily consumed placenta and amniotic fluid in addition to physically assisting the dam in the delivery of their offspring (Jones & Wynne-Edwards, 2000). Furthermore, weanling-aged male and female *P. campbelli* (average age of 24 days old) that had not been removed from their natal nest also ingested placenta and amniotic fluid while their mothers gave birth to a litter conceived during a postpartum estrus (Vella, Evans, Ng & Wynne-Edwards, 2005). Additionally, almost all 18–30 day old offspring ingested placenta in a novel chamber away from their mother, but this decreased as they reached reproductive maturity (Gregg & Wynne-Edwards, 2005). This high rate of placentophagia by juvenile *P. campbelli* does not appear to be completely due to a generalized interest in consuming novel or protein-rich foods because the juveniles were somewhat less likely to consume liver when it was presented at another time (Gregg & Wynne-Edwards, 2005).

It is unknown if placentophagia is displayed by other weanling rodents, but this may be likely in many species with a postpartum estrus that permits overlapping litters, including laboratory rats. Female rats living in non-laboratory environments can give birth to up to 5–7 litters a year and re-insemination during a postpartum estrus is common (Butler & Whelan, 1994; Davis, 1953; Davis & Hall, 1951; Leslie, Venables & Venables, 1952). This is especially true during late summer, when 30–50% of lactating female rats are also pregnant (Davis & Hall, 1951; Gilbert et al., 1980). Due to a suckling-induced delay in implantation, the birth of a subsequent litter in rats occurs approximately 25–26 days after a postpartum insemination rather than the typical ~22 days after insemination when they are not lactating (Gilbert, Pelchat & Adler, 1983; Mantalenakis and Ketchel, 1966). Weanling laboratory rats begin to independently emerge from the nest and explore an adjoining chamber when 17–19 days of age (Alberts & Leimbach, 1980), but in natural environments may not make prolonged or long-distanced excursions from the nest before 30 days of age (Calhoun, 1962), so have the opportunity to be present during the birth of younger siblings and participate in placentophagia if permitted to do so. It would be valuable to determine if weanling female rats are prone to consuming placenta because it may be a widely occurring developmental event in natural environments with significant consequences for later behavior. For example, if weanlings are similar to adult female rats, attraction to placenta or other components of the afterbirth may increase their interest in neonates and enhance the alloparental care they provide to their younger siblings. Because juvenile alloparenting increases females' later maternal responsiveness during adulthood (Gray & Chesley, 1984; Stern & Rodgers, 1988; Uriarte, Ferreira, Rosa, Seben & Lucion, 2008), there could be long-term consequences of an early attraction to afterbirth.

In the present experiments we hypothesized that if the opportunity for placentophagia during the delivery of an overlapping litter was a typical developmental event for *Rattus norvegicus* weanlings, they would display it in a laboratory environment and also show a preference for placenta even in the absence of their parturient mothers and a litter of neonates. To determine if this, we continuously recorded the behavior of groups of weanling females during their mother's delivery of a second litter that was conceived during a postpartum

estrus. We then examined if 25-day-old female rats still living with their mother and siblings would ingest placenta presented to them outside the home cage. To assess if these weanlings had a specific attraction to placenta outside the home cage, rather than just willing to eat it if it is the only option available, we allowed them to choose between rat placenta, rat liver, and a highly palatable non-protein based food (cake frosting).

Methods

Experiment 1: Weanlings' placentophagia during the birth of younger siblings

Subjects and Mating—Subjects were Long-Evans female rats born and raised in our colony, descended from rats purchased from Harlan Laboratories (Indianapolis, IN). Adult females were housed after weaning in groups of 2 or 3 same-sex animals in clear polypropylene cages (48 × 28 × 16 cm) with wood chips for bedding. Food (Tekland rat chow) and water were continuously available. The colony room was maintained at $22 \pm 1^\circ\text{C}$ and a 12:12 light/dark cycle with lights on at 0800 hr. After reaching 65 days of age, the females' estrus cycles were monitored daily with a vaginal impedance meter (Fine Science Tools, Foster City, CA) and on a day of proestrus each female was placed overnight with a sexually-experienced male from our colony. Subjects were singly housed a week before their expected date of parturition.

Starting the day before expected parturition, the presence of pups in each cage was checked twice daily and ~6 hr after parturition the dams were removed from their home cages and placed with a male overnight to be remated. Immediately after the dams were removed, their litters were culled to contain 8 pups (4 males, 4 females) and placed with surrogate lactating rats from our colony that also very recently gave birth but had their own litters removed. The following morning, the mated dams were vaginally smeared to confirm the presence of sperm and their pups were returned to them in their original home cage. The dams and litters were then left undisturbed other than for weekly cage cleaning until being videorecorded and checked for the birth of the second litter beginning 21 days later. All work was conducted in accordance with the National Institutes of Health Guide for Care and Use of Laboratory Animals and the Institutional Animal Care and Use Committee at Michigan State University.

Observation of Weanling Placentophagia—To avoid overcrowding of the home cages in anticipation of the next litter's birth, the older litters were reduced in size to contain three females when they were 21 days old. Two low-light sensitive, color video cameras were placed facing each long side of the home cage at 0900 hr and the cages recorded for 24 hr to capture the behaviors of the three weanling females in each cage ($n = 24$ weanlings in 8 litters) during the birth of the second litter. The following morning at 0900 hr, an experimenter checked for the presence of pups. If there was no second litter, the videos were deleted and the cage was recorded for another 24 hrs. Once the second litter of pups was detected, the videos from the most recent 24 hrs were analyzed. Unique patterning of the black-and-white fur on each weanling's back and skin on their tail was used to identify them on camera and the behavior of each of the three female juveniles was scored separately and continuously from the beginning of the second litter's delivery until 30 min after the last pup

was delivered. An ethogram adapted from a similar study of placentophagia by juvenile dwarf hamsters (*Phodopus campbelli*) by Jones & Wynne-Edwards (2000) was used. This included the juveniles assisting in the delivery by using paws or teeth to pull neonates from their mother's vagina, sniffing neonates, licking the neonates' skin, carrying neonates around the cage, sniffing placenta, eating part of all of a placenta, huddling over the nest while otherwise inactive, and nesting/digging. Latencies, frequencies or durations of the behaviors were recorded and both videos were consulted when a subject's behavior was obscured on one of the videos.

Data Analysis—To avoid possible litter effects complicating interpretation of the results (Abbey & Howard, 1973; Zorilla, 1997), the behavioral data were collapsed amongst responders for each behavior within each of the 8 trios of sisters to generate means and standard errors. Behavioral data were also collapsed within litters for the correlational analyses, with non-responding litters assigned zeros for each particular behavior's frequency and assigned latencies corresponding to the total duration of time they were observed. Statistical significance was indicated by $p < 0.05$.

Experiment 2: Weanlings' preference for placenta outside the home cage

Subjects—Randomly selected, unrelated female rats from our colony were used in this study when they were 25 days old ($n = 10$). This age was chosen because it was similar to the average age of the weanlings tested in Experiment 1 (see results below). The subjects remained with their mothers and natal litters until they were 21 days old, when five weanlings other than the subject and two of her female littermates were permanently removed from the home cage. Subjects then lived with their mother and two sisters until testing four days later. The mothers were not simultaneously pregnant and lactating in this experiment.

Donor Placentae and Livers—A protocol adapted from Gregg & Wynne-Edwards (2005) was used to obtain the donor tissues. Pregnant rats from the colony were euthanized with carbon dioxide on day 19 or 20 of pregnancy, when the placentae are well developed (Faria et al., 1990; Jollie, 1990). The uterine horns were rapidly removed and each fetus extracted from its amniotic sac and euthanized by decapitation. Placentae was cleared of adhering membranes but not rinsed, placed in individual plastic weighing trays, covered with Parafilm, and quickly stored at -20°C until the day of testing. Livers were also harvested from these pregnant donor females and treated similar to the placenta.

Preference Testing—Testing took place between 1200 – 1500 hr. One hour before testing, individual placentae and pieces of livers were randomly chosen, removed from the freezer and allowed to reach room temperature. A single placenta was placed on a small circular glass dish (6.35 cm diameter) and weighed (average weight of the placentae used = 460 ± 29 mg each). Pieces of a single liver and a dollop of a commercially available cake frosting that is readily ingested by pre- and peripubertal female rats (Betty Crocker Vanilla Frosting; General Mills Inc., Minneapolis, MN) (Klump, Suisman, Culbert, Kashy & Sisk, 2011) were also each placed on identical glass dishes and their weights adjusted to match the weight of the placenta within ± 10 mg.

Subjects were removed from their home cage containing their mother and two sisters and allowed to habituate for 20 min in a clean polypropylene test cage ($33 \times 38 \times 16$ cm) with wood chip bedding. After habituation, a large semi-opaque plastic funnel was placed over the weanling to help guide her to the starting position in the center of the test cage. The three dishes were then placed equidistant from each other such that one was placed along the middle of the long cage wall and the other two were placed in the opposing corners. The placement of each stimulus within the three possible cage positions was randomized for each subject. Once all three dishes were in place, the funnel was lifted and the test began. The subjects' latencies to contact, frequencies of contact, and durations of contact with each of the three plates were video recorded for 20 min and scored with a computerized data-acquisition system. Additionally, the presence or absence of upright "boxing" behavior directed at the dish of placenta was noted as a defensive behavior (Gregg & Wynne-Edwards, 2005). The dishes containing placenta, liver, and frosting were weighed immediately after testing to determine how much of each substance was consumed. Dishes were cleaned, rinsed with ethanol, and dried between uses. Subjects were euthanized with carbon dioxide after testing.

Data Analyses—Shapiro-Wilk tests confirmed that the continuous variables were normally distributed and one-way ANOVAs were then used to compare the weanlings' behaviors toward the three plates. The percentage of each substance ingested was calculated by dividing the weight ingested (pre-test weight minus post-test weight) by the pre-test weight and multiplying by 100. Two weanlings buried all three dishes with bedding by the end of their tests, which prevented an accurate measurement of how much they consumed, so they were each immediately replaced in the study by a female littermate. One other weanling was an outlier (Grubbs's test, $p < 0.02$) for the ingestion of frosting and was removed from the study. Statistical significance was indicated by $p < 0.05$.

Results

Experiment 1: Weanlings' placentophagia during the birth of younger siblings

All but one of the 24 weanling females (8 groups of three sisters) made some form of contact with placenta or neonates during the observations, and 58% of the weanlings (14/24) ate placenta (Figure 1). Except for one trio of sisters in which two females were placentophagic and one was not, either all members of a trio participated in placentophagia (4 trios) or none of them did (3 trios).

The groups of weanlings that did eat placenta began to do so with a latency of 16 ± 4 min after the start of parturition (range 8–30 min), showed 4 ± 1 bouts of the behavior (range 1–9), and did so for a total duration of 113 ± 52 sec (range 17–281 sec). Of the three trios of sisters in which none ingested placenta, some members did sniff placenta, as well as sniffed and licked the neonates, but they initiated these behaviors long after parturition had begun (> 30 min). Regardless if they ate placenta or not, weanlings who sniffed placenta did so for an average of 30 ± 11 sec, those that sniffed neonates did so for 178 ± 51 sec, and those that licked neonates did so for 76 ± 19 sec.

Other placenta- or neonate-directed behaviors were infrequently displayed. Members of only three trios huddled over pups in the nest (each for < 160 sec in total duration), three weanlings from two trios briefly carried neonates around the cage (each for < 5 sec total duration), and three weanlings from two other trios were seen to nest/dig during the delivery (each for < 50 sec total duration). No weanling assisted in the delivery of a neonate. Weanlings spent their remaining time away from the nest, most often huddled together resting or eating.

All second litters were delivered during the light photophase and the average duration of the observations (*i.e.*, starting when the first pup was expelled until 30 min after the last pup was expelled) was 139 ± 12 min. The average duration of the second pregnancy for the remated dams was 25.8 ± 0.7 days (range = 24–30 days), reflecting the delayed implantation experienced by female rats that conceive during a postpartum estrus (Mantalenakis & Kethcel, 1966; Gilbert, 1983). Although the trios of weanling females ranged in age by almost a week when their younger siblings were born, there were no significant correlations between the weanlings' ages and their latency to begin eating placenta after parturition began ($r = 0.01$, $p > 0.99$), or between their age and the frequency ($r = 0.36$, $p > 0.38$) or duration of time they spent eating placenta ($r = 0.04$, $p > 0.92$).

Experiment 2: Weanlings' preference for placenta outside the home cage

All 25-day-old females sampled each of the three options and spent an average of 168 ± 24 sec in contact with the plates or the substances on them. There were no significant differences in the weanlings' latencies to make contact with each plate (all < 50 sec; $F(2, 24) = 0.44$, $p > 0.64$), the duration of time they spent with each plate ($F(2, 24) = 0.74$, $p > 0.48$), nor their frequency of contacts with each plate ($F(2, 24) = 1.66$, $p > 0.21$) although they made contact with the plate containing placenta most often (21 ± 3 times for placenta vs. 14 ± 3 times for liver and 15 ± 2 times for frosting).

The weanlings did not completely consume any of the three substances within the 20-min tests, but consumed significantly more of the placenta and liver compared to the frosting in terms of raw weight ($F(2,24) = 4.59$, $p < 0.03$) and the proportion of the weight that had been initially provided to them ($F(2,24) = 7.22$, $p < 0.004$; Figure 2). Defensive boxing was never displayed toward any dish.

Discussion

It is well known that parturient females of most mammalian species are attracted to and ingest placenta and other components of afterbirth (Kristal et al., 2012). Interest in afterbirth by subadults has received almost no attention, but it could be present in many species that have a postpartum estrus or have a social structure involving juvenile alloparental care. We herein found that 58% of weanling female Long-Evans rats participated in placentophagia while their mothers gave birth to a subsequent litter that was conceived during a postpartum estrus. These results are consistent with those of Vella et al. (2005) who found that juvenile *Phodopus* would display midwifery behaviors, including ingestion of placenta, while their mothers gave birth. This was true for female juveniles of the monogamous and biparental Djungarian hamster (*Phodopus campbelli*), as well as female juveniles of the uniparental

Siberian hamster (*P. sungorus*) (Vella et al., 2005). Unfortunately, differences between their study and ours in the type of data collected and the data analyses prevent a direct comparison of the percentages of weanlings that were placentophages or the duration and frequency of their placentophagic behavior. Even so, one can conclude that some subadults of at least three rodent species will ingest placenta while their mothers give birth within a laboratory setting and that placentophagia by subadults is not strictly associated with the species' social organization.

While we found that most weanling female rats ate placenta during their mother's parturition, some did not. This was not related to the weanlings' age at testing, which was reasonable given that placentophagia decreases with age in *P. campbelli* (Gregg and Wynne-Edwards, 2005). Other factors distinguishing the weanling placentophages from non-placentophages could have included the behavior of their parturient mothers, and it was anecdotally noted that three of the eight dams studied lunged at, nipped, or pushed away their weanlings during parturition. A few dams were also observed to take neonates or placenta away from weanlings. Because these occurrences were rare and not exclusively seen in the cages where weanlings did not consume placenta, other influences must have been more important. One possibility is related to the fact that, with the exception of one trio, either all the sisters consumed placenta or none of them did, suggesting a genetic influence on placentophagia. There are differences between strains of nulliparous adult female mice in placentophagia (Kristal & Williams, 1973; Kristal & Eleftheriou, 1975), but if there are genetic factors influencing the likelihood of placentophagia in laboratory rats, they apparently are not heritable so unlikely to be clustered in groups of sisters (Kristal, 1980). Differences among groups of weanlings in placentophagia may instead be driven by epigenetic modifications established by the maternal behavior the entire litter received during neonatal life, which is known to alter the propensity of female rats to act maternally to neonates (Champagne, 2008) and this may extend to related stimuli including placenta or amniotic fluid. Indeed, at least some of the biological factors involved in placentophagia partly overlap with those for caregiving behaviors in maternally inexperienced female rats or hamsters (Gregg and Wynne-Edwards, 2005; Noonan & Kristal, 1979; Franz, Leo, Steuer, & Kristal, 1986; although see Kristal & Nishita, 1981).

In contrast to the mixed responding to placenta by weanlings observed in their home cages, all weanlings tested in a novel cage in Experiment 2 sampled the placenta provided (in addition to sampling the liver and frosting). Thus, any stimuli from their parturient mother and littermates are not required for weanling female rats to eat placenta. The higher propensity to ingest placenta outside the home cage could be related to the absence of interfering mothers or distracting littermates, but could also be due to the novelty of the testing procedure. Kristal, Peters, Franz, Whitney, Nishita & Steuer (1981) have suggested that parturition may act, in part, as a stressor to induce placentophagia in mother rats. This is indicated by their findings that most very late-pregnant rats will not eat donor placenta until just before or after parturition has begun, that placentophagia remains low in pregnant rats that are Cesarean-sectioned the day before parturition, but it is increased in nulliparous rats by various laboratory stressors experienced days or weeks before testing. Whether stress from acute handling and novelty experienced soon before testing can produce a similar

effect in weanling female rats remains to be experimentally determined. Another difference between the two experiments perhaps contributing to differences in the percentage of weanling females that ate placenta is that the placenta available in Experiment 1 were fresh but in Experiment 2 were frozen and thawed. If juvenile rats are able to make that distinction (see Engwall and Kristal, 1977), our results would suggest they somewhat preferred frozen and thawed over fresh placenta, although others have argued that the opposite is more likely (Gregg and Wynne-Edwards, 2005). Lastly, it was also the case that the mothers in Experiment 1 were pregnant and lactating, while those in Experiment 2 were only lactating. Differences between these dams in their maternal behavior, milk production, or milk content (Koiter, Moes, Valkhof & Wijkstra 1999; Martinez-Gomez, Juárez, Distel & Hudson 2004; Uriarte et al., 2008; Woodside, Wilson, Chee & Leon, 1981) could have contributed to differences in the placentophagic behavior of their weanling daughters (see Clark, Stiver, Teall & Galef, 2006).

All weanling females sampled placenta during our preference tests in Experiment 2, but they did not seem to be very enthusiastic placentophages, with an average of ~15% of the placenta ingested by weight. This can be contrasted with juvenile *P. campbelli*, in which almost all subjects completely consumed two full placentae during a 10-minute test (Gregg & Wynne-Edwards, 2005). This might reflect a species difference related to the biparental social structure in *P. campbelli*, such that all of its members are highly disposed to exhibit placentophagia even outside the natal nest. In support, juveniles of the uniparental *P. sungorus* are less likely to completely consume placentae in a novel testing chamber than in the natal nest with their parturient mother present (Gregg and Wynne-Edwards, 2006). It is important to note that the *Phodopus* juveniles studied by Gregg and Wynne-Edwards (2005, 2006) were not given a choice of novel substances to ingest, unlike our rats given liver and cake frosting at the same time as placenta, which divided the weanling rats' attention among multiple attractive stimuli and this could have contributed to the apparent species differences in the weanlings' appetite for placenta.

Our results from the preference tests also demonstrated that weanling female rats did not show a specific preference for placenta. They ate similar amounts of placenta and liver, and a small but significantly greater amount of both compared to frosting. Placenta and liver are more similar to each other in their higher protein content compared to the frosting (which contained no protein), and the weanlings' preference for them is consistent with the increasing protein intake compared to fat intake in weanling female rats seen in other studies (Leibowitz, Akabayashi, Alexander, Karatayev, & Chang, 2009). These results also suggest a non-specific interest in protein-rich vascularized tissue, a result that could not have been revealed during the undisturbed home cage observation when only placenta was available. This lack of preference by weanling female rats is similar to the slight and non-significant difference between ingestion of placenta and liver by juvenile female *P. campbelli* when they were offered them non-simultaneously (Gregg & Wynne-Edwards, 2005). Both cases can be contrasted to parturient placentophagic rats, which will not readily eat liver or ground beef (Kristal, 1973). In any case, a specific preference for placenta in juvenile female laboratory rats and *P. campbelli* may be unnecessary in a typical natal nest because, unless

neonates are delivered dead or killed, there would be no other non-living vascularized tissues present when the dams are giving birth.

Unlike some species including sheep (Levy & Poindron, 1987), exposure to or ingestion of placenta and amniotic fluid are not required for the onset of maternal behavior in inexperienced laboratory rats. Indeed, Cesarean-sectioned females display normal short- and long-term responses to pups (Moltz, Robbins & Parks, 1966; Bridges, 1977). Even so, smearing placenta on pups is particularly effective in shortening the latency to induce maternal behavior in nulliparous rats and hastens the onset of maternal behavior in pseudopregnant females (Kristal, Whitney & Peters, 1981; Steuer, Thompson, Doerr, Youakim & Kristal, 1987). In these cases, afterbirth intensifies females' attraction to pups, which may help them initiate physical contact with the litter and more firmly establish maternal behavior (Kristal, Whitney & Peters, 1981). In addition, consuming afterbirth permits ingestion of steroids (Strauss, Martinez, & Kiriakidou, 1996), lactogens (Lin, Poole, & Linzer, 1997), and oxytocin (Lefebvre, Giaid & Zingg, 1992). Endogenous release of these neurochemicals promotes the onset of maternal behavior (Numan, Fleming & Levy, 2006; Lonstein, Pereira, Marler & Morrell, 2013) and perhaps this also occurs after ingesting them through an exogenous source like placenta. Furthermore, afterbirth contains a substance that rapidly potentiates the opiate-mediated hypoalgesia that occurs during parturition (Kristal, Thompson, & Grishkat, 1985; Zhu & Pintar, 1998; Doerr & Kristal, 1989) and this substance simultaneously potentiates opioid activity in the ventral tegmental area to hasten the onset of maternal behavior (Neumann, Hoey, Daigler, Thompson & Kristal, 2009).

It is unknown if placentophagia has similar behavioral consequences in weanling female rats. Juvenile female rats are extremely interested in neonates and display maternal-like behaviors toward them (Mayer & Rosenblatt, 1979; Bridges, Zarrow, Goldman, & Denenberg, 1974; Brunelli, Shindledecker & Hofer, 1985). Interest in and ingestion of placenta and amniotic fluid could further increase juveniles' attraction to neonates. This would be particularly useful when mothers give birth after being inseminated during a postpartum estrus because juvenile rats' attraction to neonates sharply declines beginning around 24 days of age, which is around the age when their mothers would give birth to the second litter (Mayer & Rosenblatt, 1979; Bridges et al., 1974; Brunelli et al., 1985). Therefore, interest in afterbirth and placentophagia may prevent this decline in alloparental care, thereby increasing the weanlings' inclusive fitness. Because interacting with pups during juvenile life has a long-term positive effect on females' later responding to neonates (Gray & Chesley, 1984; Stern & Rodgers, 1988; Uriarte et al., 2008), early placentophagia could further increase fitness by improving a females' future postpartum maternal performance.

In sum, many weanling female laboratory rats will participate in placentophagia while their mothers give birth to a subsequent litter and most will ingest placenta outside of the home cage. Weanling female rats do not appear to have a specific preference for placenta, though, when given a choice among protein-based substances. Any effects of placentophagia on the brain or behavior of weanling female laboratory rats requires further study, but it could affect their alloparental and later postpartum maternal behaviors. The implications are even

broadened when one considers that the periadolescent period is a time of tremendous neurobehavioral plasticity (Andersen, 2003; Fone & Porkess, 2008; Sisk, Gore & Lonstein, 2012; Spear & Brake, 1983) and that the social and non-social stimuli weanlings receive during the birth of an overlapping litter is likely to be a striking developmental event. Nonetheless, it is an event precluded by the animal husbandry implemented in most laboratory colonies, and this presumably has implications for studying and understanding species-typical neurobehavioral development in rats and other animals with a postpartum estrus that results in overlapping litters of offspring.

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Figure 1.
Still image of a 25-day-old female rat ingesting placenta in the home cage during the birth of a litter of younger siblings. The parturient mother can be seen in the background.

Figure 2. Mean (+SEM) weight (top panel) and percentage (bottom panel) of placenta, liver and frosting ingested by 25-day-old female rats tested in a novel chamber. Different letters above bars indicates statistically significant differences between groups, $p < 0.05$.