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MATING-INDUCED REGULATION OF INFANTICIDE IN MALE MICE: FETAL PROGRAMMING OF A UNIQUE STIMULUS-RESPONSE

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Infanticide is a unique form of intraspecific aggression. Most forms of intraspecific aggression rarely lead to the death of interacting animals, whereas infanticide — by definition — is the killing of pre-weanling conspecific young. The fact that mammals of both sexes kill young of their own kind is well documented. Much of the early work on this controversial subject assumed that infanticide was maladaptive and symptomatic of grossly abnormal conditions in nature or the laboratory. Calhoun [1], in his classic studies of the Norway Rat, noted that sociopathologic conditions, such as excessive crowding, can indeed lead to a severe social breakdown and thus a high incidence of infanticide. In recent years, however, a prominent new view of infanticide has emerged: such behavior can be adaptive and routinely occurs as a well-defined behavioral strategy in a variety of mammals and other vertebrates [2]. Field studies of African Lions [3,4] and Indian Langurs [5], and laboratory studies of house mice [6,7] have dramatically documented the reproductive advantages that accrue when an infanticidal male usurps the territory of a rival. By killing infants, a conquering male eliminates his competitor's offspring and re-establishes the breeding cycle of females for his own reproductive benefit [2,8].

The male house mouse (*Mus domesticus*), in particular, has become the focus of much research regarding the neural and endocrine substrates responsible for infanticide [9]. The physiology of the male's infanticidal strategy is especially exciting, since the way a male house mouse behaves toward pups is governed by a unique stimulus-response system triggered specifically by ejaculation during coitus [10]. The act of ejaculation promotes infanticide in most male mice; however, by the time a male's own sired offspring would be born around three weeks after mating, infanticide is inhibited. When infanticide ceases, most males suddenly express parental behavior similar to a lactating female. This phenomenon also occurs in the Norway Rat [11]. Virtually all male mice and rats are thus likely to eliminate pups following mating but are inhibited from harming their own progeny during their mate's lactation. We know of no other time-dependent phenomenona in mammals where a dramatic shift in adaptive behavior can be programmed to occur weeks after a specific stimulus such as ejaculation.

While substantial differences have been noted in the background frequency of infanticide and parental behavior in males from various house mouse stocks, the mating-induced phenomenon is well-defined and remarkably consistent among both wild [7,12] and laboratory stocks [6,10,13]. The remainder of this chapter presents a brief synopsis of our current understanding of the regulation of this behavioral suite. Our experiments specifically address how baseline frequencies and temporal changes in infanticide and parental behavior occurring after mating in male mice are programmed by differential steroid exposure during fetal development. We will thus establish links between fetal development and the hormonal factors

influencing infanticide in naive and newly-mated adults and examine the inverse relationship between infanticide and intermale aggression. One of the unifying themes of this chapter concerns the evolution of unique hormone-behavior interactions correlated with the natural history of wild mouse populations. Some of our results have unusual physiological interpretations and suggest new hormonal and neural phenomena to investigate.

THE MATING-INDUCED REGULATION OF INFANTICIDE AND PARENTAL BEHAVIOR

The temporal changes we observe in the frequency of infanticide and parental behavior in males from our CF-1 laboratory stock can be partitioned into several distinct phases: 1) *Pre-mating* — where about 50% of all virgin CF-1 males exhibit spontaneous infanticide when they encounter pups while about 50% exhibit spontaneous parental behavior or they do not harm pups; 2) *Ejaculation* — where infanticide is enhanced as soon as mating occurs such that 85-90% of CF-1 males will now attack and kill pups during part or most of their mate's pregnancy; and 3) *Mid- to Late Pregnancy* — where infanticide is inhibited such that 80-90% of all CF-1 males shift to parental behavior prior to the birth of pups and are thus prevented from harming their own offspring. This unique response pattern occurs even when CF-1 males are isolated and deprived of female cohabitation immediately after mating [10]. Vom Saal [10] also identified a fourth phase: between 50 and 60 days after mating most parental CF-1 males suddenly become infanticidal again, and this shift coincides with the time when pups are normally weaned and depart from their natal environs. The studies reported here, however, focus on the responses of sexually-naive males and the changes in infanticide occurring between ejaculation and the birth of pups. The mechanisms regulating this behavioral sequence seem unprecedented in mammals, since some of the behavioral transitions are programmed to occur weeks after the stimulus of ejaculation during mating. Male mice have spontaneous ejaculations nearly every night [14], but this does not affect their behavior toward pups.

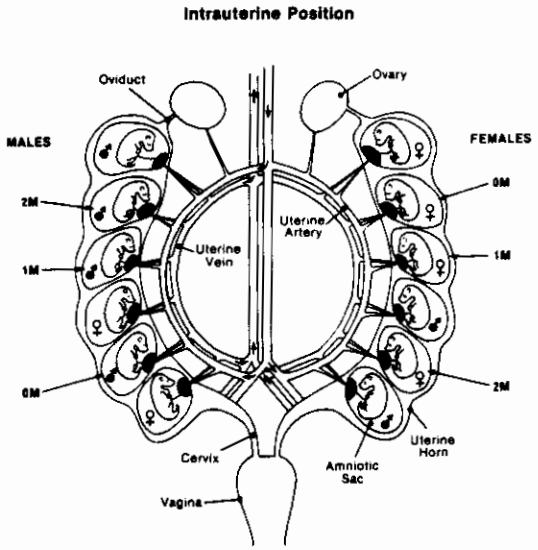

FIGURE 1. Diagram of the uterine horns and vascular supply of a pregnant mouse at term. The labels 0M, 1M and 2M refer to the number of male fetuses an individual is contiguous to: 0M= between 2 females; 1M= between a male and female; 2M= between two males.

ASSESSING HOW A MALE BEHAVES TOWARD PUPS

When a male house mouse encounters a neonate he either attempts to kill it or he does not harm it. These are clear-cut responses. We assess a male's behavior by quietly placing a 1-5 day old pup at one end of the male's home cage farthest from his nest. If a male is infanticidal, he will typically approach the pup, rattle his tail, and suddenly lunge at and kill the pup with rapid bites to the head and back. This is an acute and dramatic response, so we cannot always intervene on behalf of the pup. Wounded pups are quickly removed and humanely killed.

The opposite response from infanticide is parental behavior. Parental males groom the pup about the head and genitals and retrieve it to their nest where they incubate the pup and keep it warm. When a parental CF-1 male is allowed to incubate a pup, he appears sedated and is largely inattentive to disturbance. There is also a small subset of males (10-15%) who ignore pups, neither harming them nor exhibiting typical parental behavior [10]. When males who ignore pups are tested repeatedly, some may become infanticidal while others begin retrieving and incubating pups. Pup-ignoring males appear to straddle a neutral behavioral state between infanticide and true parental behavior. In addition, a male's reaction toward a pup seems to be a non-specific response, with no evidence for kin recognition. Previous studies have shown that neither the sex, age (1-10 days old) nor paternity of the pup has any discernable influence on a male's propensity to exhibit infanticide or parental behavior [6,7,10].

FETAL PROGRAMMING OF INFANTICIDE AND PARENTAL BEHAVIOR

Polytocous mammals such as the house mouse have multiple offspring in large litters. The intrauterine position phenomenon describes the fact that developing fetuses are positioned randomly in the uterine horns [15] and exposed to differential sex steroid concentrations in relation to whether they develop next to same sex or opposite sex fetuses [16,17]. Blood testosterone concentrations during days 17-18 of fetal development are significantly higher in fetuses that develop between two male fetuses (a 2M individual) than fetuses that develop between two female fetuses (a 0M individual). Likewise, fetuses that develop between a male and a female fetus (a 1M individual) routinely show intermediate concentrations of blood testosterone. By convention, male fetuses are the reference sex used for classifying 0M, 1M, and 2M individuals. Figure 1 shows the scheme used to classify the position of male and female fetuses in the uterine horns.

The intrauterine position phenomenon has become a powerful physiological model for examining the relationship between fetal hormone exposure and variation in adult characteristics in both sexes. An individual's intrauterine position has been correlated with a wide range of variation among reproductive, behavioral, and morphologic characteristics in adult animals [18,19]. For example, 2M males (those that develop between two male fetuses) are more aggressive toward other males than are 1M or 0M males. In general, the intrauterine position effect results in a gradient of responses with the 0M versus 2M animals exhibiting characteristics at opposite ends of this gradient. This is a reliable, naturally-occurring phenomenon. While much of the research on intrauterine position has been accomplished with laboratory stocks of house mice, position effects have been verified in wild-trapped house mice [unpublished observation], as well as rats, gerbils and pigs [20-22].

Statistically, in any large random sample of CF-1 house mice (averaging 6 pups/uterine horn), 50% of all individuals are 1M while 20% are 0M and 20% are 2M (about 10% of individuals cannot be classified by this scheme) [15]. A schematic illustrating the trimodal frequency distribution of 0M, 1M, and 2M phenotypes and their relative position along a

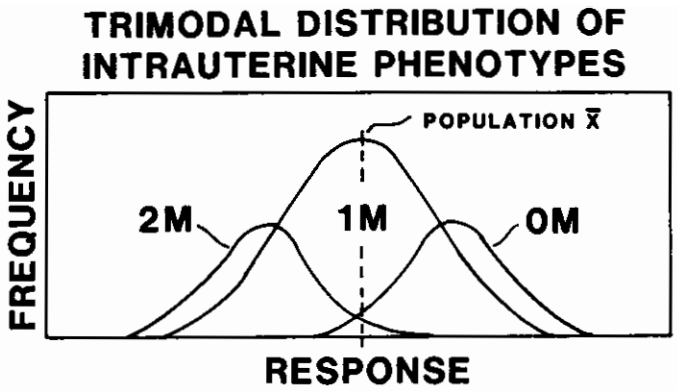

FIGURE 2. Frequency distribution and generic response pattern of 2M, 1M and 0M individuals in a large population sample.

generic response gradient is shown in Figure 2. One point needing emphasis here is that, like fetal concentrations of blood testosterone, whenever a correlation has been found relating a particular behavioral, somatic or reproductive characteristic to prior intrauterine position, 1M individuals have, on average, always been found intermediate in their characteristics between the 0M and 2M extremes. Thus, the population mean is centered on 1M individuals.

As noted earlier, virgin CF-1 males tested in our laboratory always yield a distribution of about 50% spontaneously infanticidal versus 50% spontaneously parental or pup-ignoring males. Vom Saal [23] noted that when virgin 1M males were tested for their behavior toward young, half were infanticidal and half were parental or ignored a pup. This finding matches the predictions of the model in Figure 2. Given the ability of intrauterine positioning to reliably predict profound differences in later adult characteristics, we suspected that individual variation among males in infanticide and parental behavior as a result of mating might, like many other characteristics of house mice, also be programmed during fetal development.

In a recent experiment [unpublished] we compared the mating-induced responses of virgin males obtained from 0M versus 2M intrauterine positions (Since 1M individuals have always been found intermediate in their characteristics, ethical considerations did not justify the use more animals than necessary to test our hypothesis). To obtain males from known positions *in utero*, pregnant CF-1 females were sacrificed by decapitation on Day 19 of pregnancy (insemination = Day 0) several hours before the time of normal parturition. Pups were removed sequentially from the uterine horns by cesarian section and classified in accordance with the scheme in Figure 1. Male 2M and 0M pups were raised in randomly mixed litters by foster females.

When adult (90 days of age), 19 0M and 15 2M males were each tested with a newborn pup at three time points: several days before mating, one day after mating, and at 21 days after mating. As shown in Figure 3, sexually-naive 0M males were more than twice as likely to exhibit infanticide than their 2M counterparts: 63% versus 27%, respectively ($p < .05$). This replicates differences previously noted between virgin 0M and 2M males [23]. At one day after mating, infanticide was elevated in both the 0M and 2M groups; several parental males became infanticidal as a result of the ejaculation stimulus. However, by three weeks after mating (when a male's pups are born), 100% of the 2M males had shifted to parental behavior, whereas 37% of the 0M males still exhibited infanticide ($p<.05$). Thus, some males who develop *in utero*

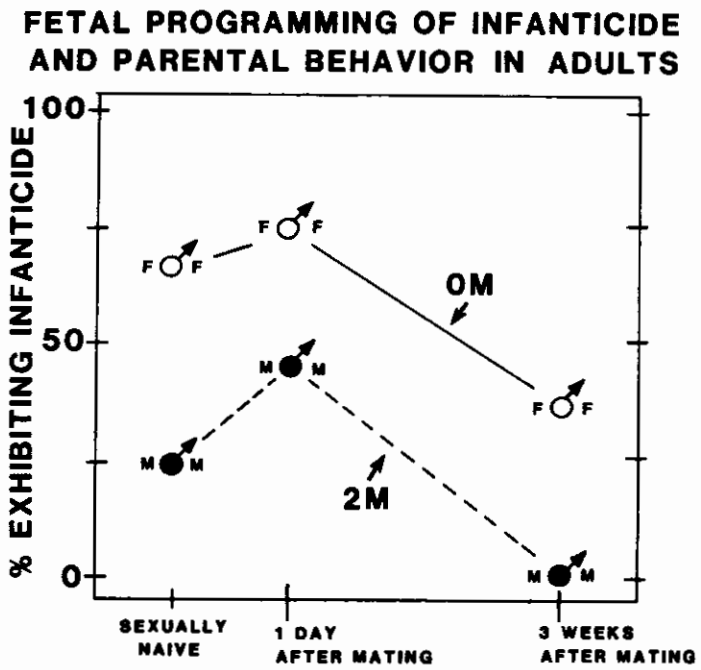

FIGURE 3. Percentage of 2M versus 0M males exhibiting infanticide before mating, one day after mating and three weeks after mating [unpublished observation].

between 2 female fetuses (0M) are rendered insensitive to the inhibitory effects of mating on infanticide. A primary conclusion here is that the suite of behaviors involved in the mating-induced regulation of infanticide, like many other characteristics studied in house mice, is indeed influenced by whether an individual develops in utero next to same or opposite sex fetuses.

PROGRAMMING OF BEHAVIORAL THRESHOLDS DURING FETAL DEVELOPMENT

Because the adult responses of 2M versus 0M individuals are a consequence of being at opposite ends of a gradient of sex steroid exposure during late fetal development, a second unique set of conclusions can be generated about the temporal dynamics of infanticide and parental behavior. Our evidence suggests adult male mice are programmed for different behavioral thresholds as a function of intrauterine position. Figure 4 represents a behavioral matrix illustrating our suggested scheme for relating increasing fetal testosterone exposure to one of four possible behavioral sequences exhibited by CF-1 males. The temporal changes in infanticidal and parental strategies are depicted here in three phases: 1) *Pre-mating*, when males are virgin; 2) *Pregnancy*, defined as the time from ejaculation until a mate gives birth; and 3) *Lactation*, when a male's own sired pups are present and vulnerable to attack. Virgin males can begin their behavioral sequence as either parental or infanticidal. When spontaneously parental males are mated, some will remain parental, while others will be stimulated to exhibit infanticide as a result of ejaculation. Spontaneously infanticidal males, however, always remain infanticidal after ejaculation [10].

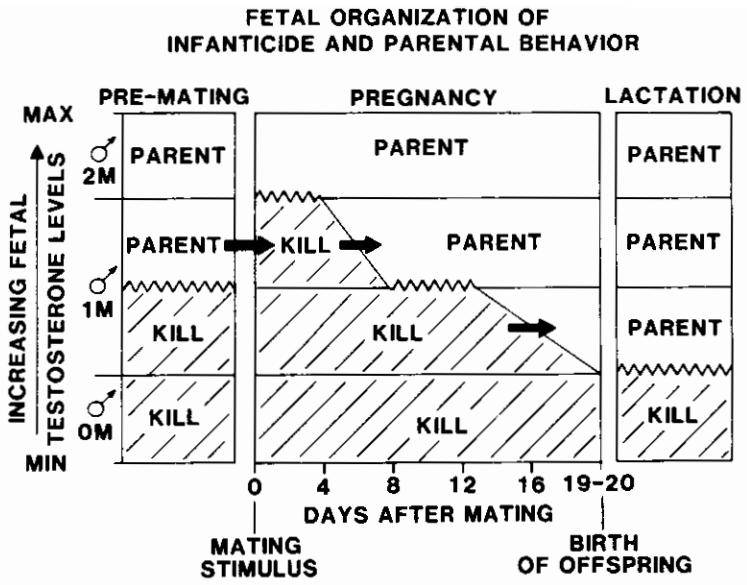

FIGURE 4. Behavioral matrix relating increasing fetal testosterone exposure to the temporal changes in infanticide and parental behavior exhibited by adult CF-1 males.

2M males receive maximal testosterone exposure during late fetal development. They are the most likely individuals to exhibit a "pure" parental strategy. In sharp contrast, 0M males receive minimal testosterone exposure during late fetal development, and they are the most likely individuals to exhibit a "pure" infanticidal strategy. We have observed that about 10-20% of CF-1 males are always parental and about 10-20% of males are always infanticidal, regardless of their mating history, while 60-80% of all males exhibit either a *PARENT* to *KILL* to *PARENT* sequence or a *KILL, KILL* to *PARENT* sequence during the respective pre-mating, pregnancy and lactation phases. The frequencies of these four strategies match remarkably well, percentagewise, with the expected proportions of 2M, 1M and 0M phenotypes. Since there is substantial overlap in response characteristics between 2M and 1M adults and between 1M and 0M adults (see Figure 2), one concept concerning Figure 4 needs to be explicitly emphasized: our scheme does not imply that each of the three intrauterine phenotypes is locked-in to one of the specific behavioral patterns shown here. Like Figure 2, this diagram is only meant to visualize a probability distribution showing which type of adult is most likely to exhibit spontaneous infanticide or parental behavior, and which type of adult is most likely to follow through on a specific behavioral sequence after mating.

A second point evident in Figure 4 is that in previous studies where spontaneously parental CF-1 males became infanticidal as a result of ejaculation, most of them re-expressed their parental behavior within 8 days. Spontaneously infanticidal CF-1 males, on the other hand, typically do not cease killing pups and express parental behavior until 10 or more days after they mate [10]. This suggests that fetal exposure to sex steroids also influences the timing of the inhibition of infanticide following ejaculation.

THE EFFECTS OF GONADAL AND PITUITARY HORMONES IN ADULT MALES

Differential exposure to sex steroids during fetal development clearly influences the way adult male mice behave toward young. Some steroid-sensitive behaviors are "organized"

during perinatal development and do not require the presence of specific gonadal hormones in order for the behavior to occur in adulthood [24,25]. But other behaviors may be "sensitized" during perinatal development and therefore require the presence of gonadal hormones in adulthood for the behavior to occur ("activation").

When virgin CF-1 males are castrated and allowed to interact with a pup for the first time, all males behave parentally. If, however, these same castrated males are implanted with a 1 cm silastic capsule containing 5 mg of crystalline testosterone (dissolved in .02 cc of sesame oil) and retested for their behavior toward pups several days later, half of the previously parental males will now exhibit infanticide. This finding mimics the typical 50/50 proportion of spontaneously infanticidal versus non-infanticidal males observed when intact CF-1 males are tested. Removal of the testosterone capsule abolishes infanticide. Concurrent exposure to testosterone thus seems required for a virgin male to exhibit infanticide when he encounters a pup [23].

The act of mating causes a dramatic surge in LH (luteinizing hormone) and testosterone in male mice [26], so we tested whether the hormonal changes associated with mating might be responsible for mediating changes in a male's behavior toward pups. Twenty-two spontaneously infanticidal males were hypophysectomized and castrated, and, in order to maintain both their ability to mate and exhibit infanticide [23], they were also implanted with a 5 mg testosterone capsule. Half of the males were allowed to mate. When tested with a pup at 20 days after mating, only 1 out of 12 mated males exhibited infanticide while 6 out of 10 non-mated males still exhibited infanticide ($p<.05$; Figure 5) [27]. Hypophysectomized, mated males thus showed a post-mating inhibition pattern identical to that observed in previous experiments using intact males [10]. This finding reveals that the mating-induced inhibition of infanticide is a neurally-timed and mediated response that operates independently from pituitary hormone secretions or *changes* in gonadal secretions as a result of mating. However, because castrated males must be given replacement testosterone to accomplish mating, testosterone may be a permissive factor in initiating this response sequence.

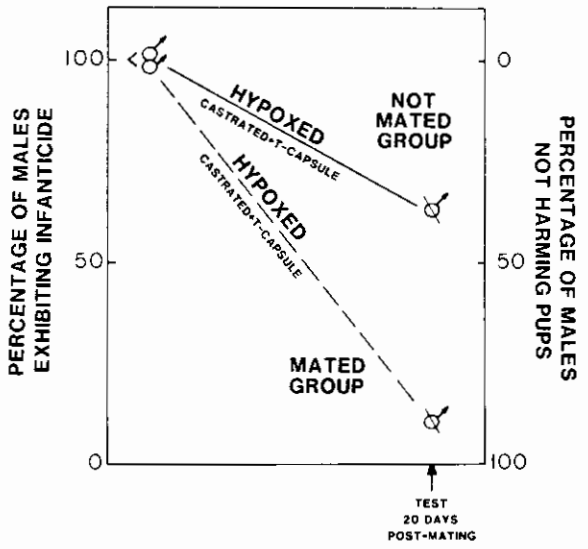

FIGURE 5. The Effects of hypophysectomy on the mating-induced inhibition of infanticide. Males were also castrated and implanted with a testosterone capsule (to maintain mating behavior and infanticide). At 20 days after mating, there was a significant inhibition of infanticide in the mated group compared to the non-mated group ($p<.05$).

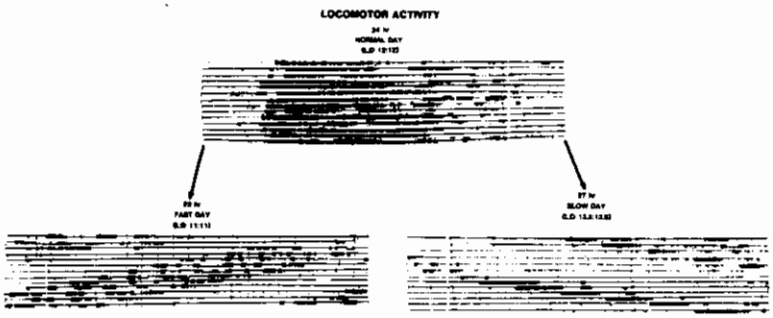

FIGURE 6. Daily locomotor patterns in a representative *fast day* male (left) and *slow day* male (right); 24 hr strips from an event recorder are pasted consecutively over several days. The dark bars in each strip represent when the animal was active on a running wheel. All individuals began their entrainment at a normal L:D 12:12 cycle; the top picture represents a typical activity pattern of a standard 24 hr day. The stair-step patterns generated in the lower pictures demonstrate that the *fast day* animal was phase-advanced by 2 hrs each day while the *slow day* animal was phase-delayed by 3 hrs each day.

HOW DO MATED MALES KEEP TRACK OF TIME?

The stimulus of ejaculation results in an unusually prolonged sequence of behavioral changes. A mated male must somehow track his mate's pregnancy and be able to stop killing pups and behave parentally at the appropriate time. This prompted us to ask how mated males keep track of time. Since CF-1 males do not need female cues to exhibit this response [10], this clearly suggested the efficacy of this strategy depended on a unique biological timekeeper. We speculated that mated males could keep track of time either 1) by measuring the absolute amount of time passing after ejaculation, or 2) by counting the number of light/dark cycles experienced after ejaculation. Photoperiodic variation in nature always provides infallible temporal cues for entraining daily (circadian) and seasonal cycles of feeding, breeding, metabolism and movement [28,29], so we suspected the latter hypothesis. To test both possibilities, we used an experimental paradigm that allowed us to distinguish between absolute time (a standard 24 hr day) versus the number of light/dark cycles experienced: CF-1 males were thus housed at artificially *fast* (L:D 11:11 = 22 hr.) versus artificially *slow* (L:D 13.5:13.5 = 27 hr) light/dark cycles.

One hundred adult males were placed in light-tight, coffin-sized boxes illuminated inside with a 15 w fluorescent lamp (L:D 12:12 starting light/dark cycle). Fifty males in each group were slowly adapted over a 25 day period to the 22-hour *fast day* cycle or the 27 hr *slow day* cycle by either increasing or decreasing the length of their light and dark exposure by 2.5 to 3.5 minutes each day. To verify behavioral entrainment, the locomotor patterns of several randomly chosen males in each group were monitored in cages with a running wheel interfaced to an event recorder (Figure 6). Males were allowed to mate and then screened for infanticide 1 day after ejaculation. Parental males were discarded from the experiment while the remaining infanticidal males (about 85% in both the *fast* and *slow day* groups) were retested with a pup between 16 and 25 absolute (24 hr) days after mating. The rationale behind our test procedure is illustrated by the "phase timeline" in Figure 7. Specifically, half of the *fast day* males were retested at 16.3 absolute days (=18 light/dark cycles) and half were retested at 20 absolute days

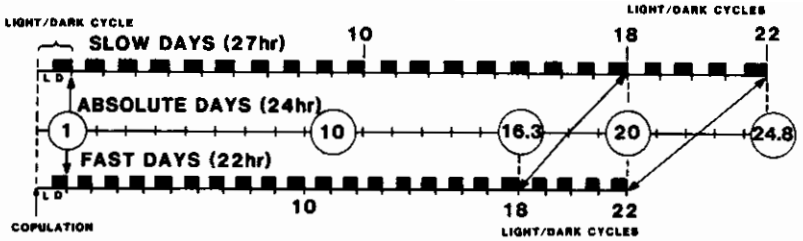

FIGURE 7. The relationship between absolute time (24 hr days) and the increasing desynchronization of light/dark cycles experienced by both groups during the course of this experiment. The alternating dark bars on the *fast* and *slow* time scales represent the dark phase of the repeating light/dark cycle. By 20 absolute days after mating, *fast day* males had experienced 4 more light/dark cycles than *slow day* males.

(=22 light/dark cycles) after mating, while half of the *slow day* males were retested at 20 absolute days (=18 light/dark cycles) and half were retested at 24.8 absolute days (=22 light/dark cycles) after mating. Our objective here was to directly compare both groups at 20 absolute days after mating and also control for the number of equivalent light/dark cycles experienced by both groups (18 versus 22 cycles).

Figure 8 shows the results. The post-mating inhibition of infanticide is graphed here in two complementary perspectives: first, in relation to the number of absolute (24 hr) days experienced after mating, versus second, in relation to the number of light/dark cycles experienced after mating. When viewed side-by-side, the graphs suggest the presence of a unique biological timekeeper. At 20 absolute (24 hr) days after mating there was a significant difference in the frequency of infanticide between the *fast* and *slow day* groups (13% versus 61%, respectively; $p < .005$), suggesting that mated males do not use absolute time after mating as a cue to trigger the inhibition of infanticide [27]. Furthermore, no differences were noted in the frequency of infanticide with both groups matched for experiencing the same

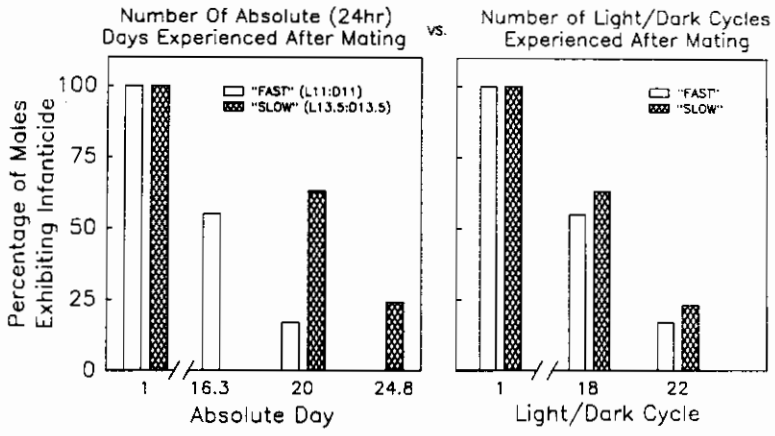

FIGURE 8. The percentage of male mice maintained at fast (=22 hr) versus slow (=27 hr) days exhibiting infanticide when graphed in relation to absolute (=24 hr) days versus the number of light/dark cycles experienced after ejaculation.

number of light/dark cycles. Male house mice thus apparently measured the passage of time by "counting" the number of light/dark cycles they experienced after mating. Our findings suggest that up to three weeks after the specific stimulus of coital ejaculation, a physiological mechanism that somehow assesses light/dark cycles can trigger the inhibition of infanticide and the onset of parental behavior.

The fact that dramatic shifts from infanticide to parental behavior in male mice parallel the behavioral and temporal dimensions of pregnancy in females is in itself interesting. Infanticide is also a fundamental component of the behavioral repertoire of female house mice — virtually all pregnant wild-stock females kill pups up to the time of parturition, at which time they become parental [12,30]. Thus, both sexes exhibit infanticide and seem to share a common suite of parental behaviors expressed at the time pups are born. But female house mice rely on the cues from developing fetuses and always gauge the length of pregnancy in absolute time. Even when entrained to extreme light/dark cycles mimicking a 20 versus 28 hour day, female house mice still give birth the same number of absolute days after insemination [31].

Male mice have apparently co-evolved a novel timekeeping solution for synchronizing their behavior toward pups with the duration of their mate's pregnancy. Some photoperiodically-mediated phenomena in rodents, such as the preovulatory surge in LH, and hence, the organization of estrous cycles, regularly occur at 4-5 day multiples of daily light/dark cycles [32]. Unlike our present finding, however, these events are mediated by cyclic changes in the secretion of pituitary and gonadal hormones. As described in the previous section, the mating-induced shift from infanticide to parental behavior occurs in male mice even in the absence of the pituitary or changes in gonadal hormones. Our experiment suggests that a neural signal can be propagated through time for many days by coupling the signal with a neural mechanism which "counts" days, where days are defined by repeating light/dark cycles. The 2 to 3 week timespan intervening between copulation and the inhibition of infanticide (and onset of parental behavior) redefines the possible temporal and behavioral relationships between a neural stimulus and its response.

This unique stimulus-response system seems to provide most mated males with a fail-safe mechanism for ensuring the inhibition of infanticide by the time pups are born. But in typical post-mating situations, redundant cues from a female may also facilitate a male's response. Some debate has been generated over the relative importance of ejaculation versus female cohabitation in inhibiting male infanticide. Recent work from other laboratories, however, has clearly demonstrated that ejaculation and female cohabitation are both effective in this regard [12,13]. While ejaculation alone can inhibit infanticide in male rats, long-term exposure to just the soiled bedding of a pregnant female is also an effective means of inhibiting infanticide in sexually-naive males [11].

These results are not surprising, since chemical cues are the overwhelmingly dominant mode of communication among rodents. The multitude of complex chemosensory cues mediating reproduction in male and female rodents is widely known, and pheromonally-induced responses are usually augmented by tactile and other multisensory cues [33]. Although we have not tested this possibility, our CF-1 males may also exhibit a sensitivity to such cues. As demonstrated earlier, the act of mating does not seem to inhibit infanticide in many 0M male mice — it is possible that these individuals need to cohabit with a female before they can exhibit parental behavior. In any case, the underlying neural mechanism which inhibits infanticide after mating in most CF-1 male mice appears strong enough to override or mask whatever extrinsic chemical and tactile cues might also be operating to modify their behavior toward young.

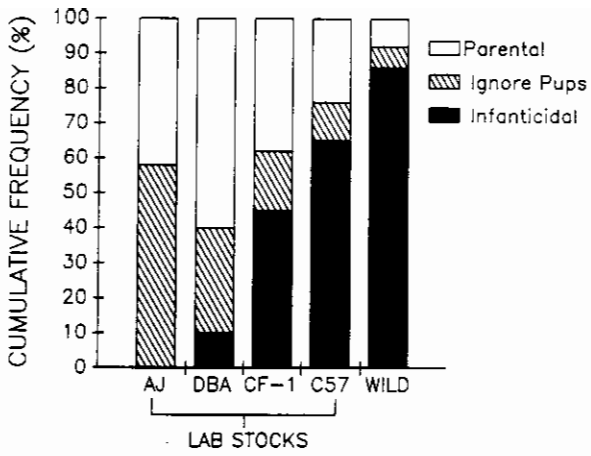

FIGURE 9. **Variation in spontaneous infanticide and parental behavior among several stocks of house mice.**

DIFFERENCES AMONG LABORATORY AND WILD STOCKS OF HOUSE MICE

So far, the focus of this chapter has been on experimentation with CF-1 stock males from our laboratory colony. Nevertheless, there is substantial variation in the frequency of spontaneous infanticide and parental behavior among mouse stocks, and this variation provides a useful comparative tool for understanding what dimensions of behavior are genetically malleable and what sort of influences fetal hormonal exposure has upon adult responses toward young.

Intense and often unplanned artificial selection in the laboratory can rapidly change the physiology and behavior of house mouse stocks [34]. Figure 9 compares the widely-different background frequencies of spontaneous infanticidal and non-infanticidal behavior observed among males of four laboratory stocks and a wild-trapped stock from Columbia, Missouri. An interaction between the length of a female's pregnancy and a stock's sensitivity to sex steroids may account for some of the variation in spontaneous infanticide among mouse stocks [35]. The duration of pregnancy can vary and thus lengthen the amount of time fetuses are exposed to sex steroids *in utero*. Ancestral wild stocks, for example, usually deliver on Day 18 of gestation (insemination = Day 0), while the CF-1 stock delivers on Day 19. Wild stocks are thus typically exposed for one less day to the hormonal influences of the placenta and their fetal neighbors than their CF-1 domestic counterparts. Because testosterone exposure drops dramatically after birth in male house mice [36], one would predict an elevation of infanticidal behavior in wild stocks (see also Figure 10). Indeed, 90% of male wild mice from Missouri are spontaneously infanticidal. C57 stocks give birth on Day 18 and they also show a greater frequency of infanticide than the CF-1 stock [9]. In sharp contrast, AJ mice are a unique stock that delivers between day 20 and 21 of pregnancy. Since fetal AJ mice undergo prolonged *in utero* exposure to sex steroids, the pregnancy-length model predicts infanticide should be greatly diminished; in fact, AJ mice do not seem to exhibit spontaneous infanticide at all [unpublished]. But the length of pregnancy is not always correlated with the behavior of males toward pups. The DBA inbred line, for example, has a 19 day pregnancy and very few males are infanticidal [9].

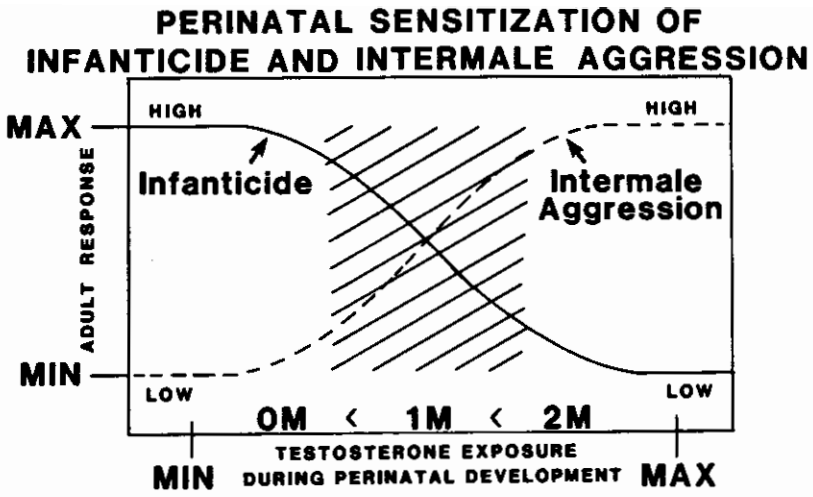

FIGURE 10. The inverse relationship between infanticide and intermale aggression in adult mice in relation to intrauterine position and fetal testosterone exposure.

Some laboratory stocks may also differ in their sensitivity to steroids due to variation in steroid production or binding [9]; thus, the quality of steroid-related responses has apparently undergone genetic changes in some inbred lines. An interesting comparison here is that in at least one inbred line (CBA mice), copulation *per se* does not seem to inhibit infanticide, whereas female cohabitation does [37]. This raises the possibility that a genetically-based shift in steroid-related responses may have rendered the mating-induced response of CBA stock males phenotypically similar to that observed in many of the 0M male phenotypes in our CF-1 colony.

The hypothesis that laboratory selection has changed the responses of various mouse stocks toward young suggests there is a great deal of flexibility in the mechanisms regulating infanticide. But at present, the mechanisms underlying stock variation remain unknown. In any case, there is enough variability and flexibility in infanticide among mouse stocks to allow a wealth of unique experimentation tracing the specific neural and endocrine pathways responsible for this behavioral suite. CF-1 males, however, seem to be a fortuitous choice for studying hormonal influences and the fetal programming of infanticide and parental behavior. First of all, CF-1 males exhibit a robust and reliable mating-induced response without the confounding factors of female cohabitation or exposure to their odors; and secondly, this stock has an intermediate gestation length and exhibits equal proportions of spontaneously infanticidal versus parental males (see Figure 9). If examined side-by-side with studies of genotype effects on infanticide in other stocks, CF-1 mice could provide a unique model for understanding how complex behaviors are switched on during fetal development, how steroids bias the neural network in one way or the other, and what types of extrinsic cueing systems also modulate infanticide and parental behavior, but without the confounding problems of systematic genetic variation.

HOUSE MOUSE SOCIAL ORGANIZATION AND THE RELATIONSHIP BETWEEN INFANTICIDE AND INTERMALE AGGRESSION

Experiments involving pharmacological manipulations have shown that exposure to elevated levels of testosterone during late fetal development greatly diminishes infanticide in male house mice — but just the opposite is true of intermale aggression [35,38]. Infanticide and intermale aggression are thus negatively-correlated (Figure 10). 2M males are more aggressive toward other males, presumably as a result of perinatal exposure to high testosterone concentrations relative to 0M males [17]. Yet, a high proportion of 2M males do not exhibit infanticide, whereas most 0M males do exhibit infanticide [23].

Vom Saal [35] noted that since high testosterone exposure during fetal life has opposite effects on adult intermale aggression and infanticide, the inverse relationship between these two types of aggression suggests they are correlated behaviors. These results concerning infanticide may, offhand, seem counter to the classical sensitizing effects of testosterone during fetal life and its role in maintaining and potentiating aggression during adulthood, but an overview of the social structure of wild mouse populations offers a parsimonious explanation of how this unique behavioral dichotomy evolved.

In free-living situations, a dominant male usually defends a territory containing several breeding females. The structure of a typical house mouse deme is illustrated in Figure 11. A 2M male may be more likely to hold a territory by aggressively repelling or subordinating rival males [19], and there is evidence that virtually all offspring in a mouse deme are produced by the one dominant male [39]. A predisposition for parental behavior would prevent a dominant 2M male from accidentally killing his own offspring. On the other hand, if a subordinate 0M male can supplant a dominant male and obtain mating, a strong infanticidal tendency would ensure the elimination of his competitor's pups. Mating does not seem to inhibit infanticide in many 0M male mice, but, as suggested earlier, these individuals may become parental before the birth of their progeny, provided they cohabit with a female. When viewed *in toto*, the natural history background and social organization of feral populations suggests that infanticidal behavior in male mice, and possibly other mammals, co-evolved in harmony with other hormone-mediated forms of aggression. Evolution has probably economized here: selection pressures have simply driven the physiological consequences of fetal testosterone exposure on intermale aggression and infanticidal behavior in totally opposite directions.

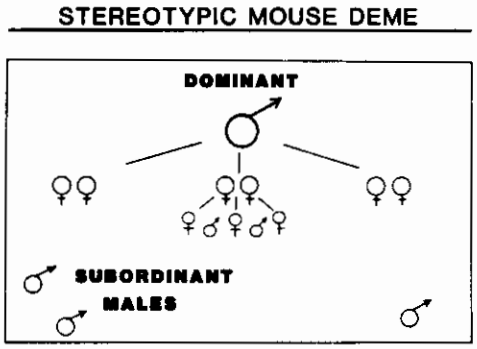

FIGURE 11. Social organization of a typical feral mouse deme.

It must be stressed, however, that while an individual's background tendency to exhibit infanticide or intermale aggression may, to a great extent, be biased during fetal development, moment-to-moment experiential factors can also play a pivotal role in regulating a male mouse's infanticidal strategy. Intense fighting, such as that which occurs when an intruding male attempts to take over another's territory, appears to potentiate infanticide in the intruder if he defeats and subordinates his opponent. In contrast, subordination in this type of situation seems to inhibit infanticide [6,13,40].

Finally, when infanticide is triggered by ejaculation in spontaneously parental males, they begin killing pups as soon as they have mated [unpublished]. This dramatic behavioral shift undoubtedly evolved in response to the reproductive physiology of the female. Female house mice enter into a strong postpartum estrous within 24 hours after parturition; thus, a spontaneously parental virgin male obtaining first mating with a postpartum female would benefit from immediately destroying her litter. Lactation can delay the implantation of fertilized eggs for a week or more, but once the suckling stimulus is removed from a newly-lactating female, the new stud male's embryos will implant immediately [6]. Even if a male mouse already has a tendency to be infanticidal before he mates, ejaculation may intensify a male's motivation to seek out and kill conspecific pups. Small mammals such as the house mouse are extremely short-lived, and a shortened generation time can have enormous cumulative benefits for the reproductive success of an individual. Infanticide in the male house mouse is a violent but extremely effective strategy for obtaining reproductive success.

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REFERENCES

1. Calhoun, J.B. (1962). Population density and social pathology. *Sci. Amer.* 206:139-148.
2. Hausfater, G. and S. Blaffer Hrdy, Eds (1984). Infanticide: Comparative and Evolutionary Perspectives. Chicago: Aldine Publishing.
3. Schaller, G. (1972). The Serengeti Lion. Chicago: Chicago University Press.
4. Packer, C. and S. Pusey (1984). Infanticide in carnivores. In: G. Hausfater. and S. Blaffer Hrdy, Eds., Infanticide: Comparative and Evolutionary Perspectives, pp 31-42, Chicago: Aldine Publishing.
5. Hrdy, S. Blaffer (1974). Male-male competition and infanticide among the langurs (*Presbytis entellus*) of Abu, Rajasthan. *Folia Primatologica* 22:19-58.
6. vom Saal, F.S. and L. Howard (1982). The regulation of infanticide and parental behavior: Implications for reproductive success in male mice. *Science* 215:1270-1272.
7. McCarthy, M. and F.S. vom Saal (1986). Inhibition of infanticide after mating in wild male house mice. *Physiol. Behav.* 36:203-209.
8. Hrdy, S. Blaffer (1979). Infanticide among animals: A review, classification, and examination of the implications for the reproductive strategies of females. *Ethology and Sociobiology* 1:13-40.
9. Svare, B., C. Kinsley, M. Mann and J. Broida (1984). Infanticide: Accounting for genetic variation. *Physiol. Behav.* 33:137-152.

10. vom Saal, F.S. (1985). Time-contingent change in infanticide and parental behavior induced by ejaculation in male mice. *Physiol. Behav.* 34:7-15.
11. Mannella, J. and H. Moltz (1988). Infanticide in rats: Male strategy and female counter-strategy. *Physiol. Behav.* 42:19-31.
12. Soroker, V. and J. Terkel (1988). Changes in incidence of infanticidal and parental responses during the reproductive cycle in male and female wild mice Mus musculus. *Anim. Behav.* 36:1275-1281.
13. Elwood, R.W. (1986). What makes male mice paternal? *Behav. Neur. Biol.* 46:54-63.
14. Huber, M. and F.H. Bronson (1980). Social modulation of spontaneous ejaculation in the mouse. *Behav. Neural Biol.* 29:390-393.
15. vom Saal, F.S. (1981). Variation in phenotype due to random intrauterine positioning of male and female fetuses in rodents. *J. Reprod. Fert.* 62:633-650.
16. vom Saal, F.S. and F.H. Bronson (1980). Sexual characteristics of adult female mice are correlated with their blood testosterone levels during perinatal development. *Science* 208:597-599.
17. vom Saal, F.S., W. Grant, C. McMullen and K. Laves (1983). High fetal estrogen titers correlate with enhanced sexual performance and decreased aggression in male mice. *Science* 220:1306-1309.
18. vom Saal, F.S. (1983). The interaction of circulating estrogens and androgens in regulating mammalian sexual differentiation. In: J. Balthazart, E. Prove and R. Giles, Eds., Hormones and Behavior in Higher Vertebrates, pp 154-179, Berlin: Springer-Verlag.
19. vom Saal, F.S. (1984). The intrauterine position phenomenon: Effects on physiology, aggressive behavior and population dynamics in house mice. In: K. Flannelly, R. Blanchard, and D. Blanchard, Eds., Biological Perspectives on Aggression, pp 135-179, New York: A.R. Liss, Inc.
20. Clemens, L., B. Gladue and L. Coniglio (1978). Prenatal endogenous androgenic influences on masculine sexual behavior and genital morphology in male and female rats. *Horm. Behav.* 10:40-53.
21. Clark, M., and B. Galef (1988). Effects of uterine position on rate of sexual development in female Mongolian gerbils. *Physiol. Behav.* 42:15-18.
22. Rhode Parfet, K., W. Lamberson, A. Rieke, F.S. vom Saal and B.N. Day (1988). Effects of intrauterine position on sexual behavior of boars. Abstract for Conference on Reproductive Behavior: Omaha, Nebraska (June 1988).
23. vom Saal, F.S. (1983). Variation in infanticide and parental behavior in male mice due to prior intrauterine proximity to female fetuses: Elimination by prenatal stress. *Physiol. Behav.* 30:675-681.
24. Beatty, W. (1979). Gonadal hormones and sex differences in nonreproductive behaviors in rodents: Organizational and activational influences. *Horm. Behav.* 12:112-163.
25. vom Saal, F.S. (1983). Models of early hormonal effects on intrasex aggression in mice. In: B. Svare, Ed., Hormones and Aggressive Behavior, pp 197-222, New York: Plenum Publishing.
26. Coquelin, A. and F.H. Bronson (1980). Secretion of luteinizing hormone in male mice: Factors that influence release during sexual encounters. *Endocrinol.* 106:1224-1229.
27. Perrigo, G., F.S. vom Saal and W.C. Bryant (1988). Changes in behavior of male mice toward young after mating are independent of changes in pituitary and gonadal hormones and occur as a function of the number of photoperiods rather than absolute time. Abstract for Conference on Reproductive Behavior: Omaha, Nebraska (June 1988).

28. Follett, B. and D. Follett, Eds. (1981). Biological Clocks in Seasonal Reproductive Cycles, Bristol: Wright Publishing.
29. Turek, F. and E. Van Cauter (1988). Rhythms in reproduction. In: J. Knobil and J. Neill, Eds., The Physiology of Reproduction, pp 1789-1830, New York: Raven Press.
30. McCarthy, M. and F.S. vom Saal (1985). The influence of reproductive state on infanticide by wild female house mice (*Mus musculus*). *Physiol. Behav.* 35:843-849
31. Davis, F. and M. Menaker (1981). Development of mouse circadian pacemaker: independence from environmental cycles. *J. Comp. Physiol.* 143(A): 527-539.
32. Fitzgerald, K. and I. Zucker (1976). Circadian organization of the estrous cycle of the golden hamster. *Proc. Natl. Acad. Sci. USA* 73:2923-2927.
33. Perrigo, G. and F.H. Bronson (1983). Communication disparities among genetically-diverging populations of deermice. In: R. Silverstein and D. Muller-Schwarze, Eds., Chemical Signals in Vertebrates: III, pp 195-210, New York: Plenum Publishing.
34. Bronson, F.H. (1984). Energy allocation and reproductive development in wild and domestic house mice. *Biol. Reprod.* 31:83-88.
35. vom Saal, F.S. (1988). Perinatal testosterone exposure has opposite effects on adult intermale aggression and infanticide in mice. In: P. Brain and S. Parmigiani, Eds., House Mouse Aggression, London: Gordon and Breach Science Publishers.
36. Pointes, G., M. Latreille, T. Mignot, Y. Janssens and L. Cedard (1979). Regulation of testosterone synthesis in the fetal mouse testis. *J. Steroid Biochem.* 11:1609-1612.
37. Kennedy, H. and R. Elwood (1988). Strain differences in the inhibition of infanticide in male mice (*Mus musculus*). *Behav. Neur. Biol.*, in press.
38. vom Saal, F.S. (1979). Prenatal exposure to androgen influences morphology and aggressive behavior of male and female mice. *Horm. Behav.* 12:1-11.
39. DeFries, J. and G. McClearn (1972). Social dominance and Darwinian fitness in the laboratory mouse. *Amer. Nat.* 104:408.
40. Huck, U., R. Soltis and C. Coopersmith (1982). Infanticide in male laboratory mice: effects of social status, prior sexual experience, and basis for social discrimination between related and unrelated young. *Anim. Behav.* 30:1158-1165.