does not seem to be possible even if it were advantageous.

The original explanation for the evolution of sex ratio (*5*) included the idea that females in better than average condition would invest preferentially in male offspring. The mothers in our colony seemed to be in excellent condition, yet control litters show sex ratios of 1.0. A proximate explanation for this is that all of the offspring born to control mothers could be weaned in excellent condition; under unrestricted circumstances, there is no reason to sacrifice any young. Behavioral sex-biased litter reduction, then, should only work in one direction, in this case against males. Brood reduction strategies seem to be a way of responding adaptively to food levels that fluctuate unpredictably over the time interval of a reproductive event.

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13. The use of this estimate of investment requires the assumption that the energy equivalent (mass-specific tissue) of the food-restricted young is the same as that of the unrestricted young. If this is not correct, the energy equivalent for food-restricted young would probably be lower than that of controls, with the result that this estimate is conservative. The actual total energy investment of mothers in young also includes the energy used by the metabolism of young, but I have not included that component, since I have not measured metabolic rates of food-restricted young; small size and cold bodies indicate that the metabolic rate of males would probably be less than that of females. This should also reinforce the conclusion from these estimates that food-restricted females switch their investment of energy to favor females.
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## Ascending Endorphin Inhibition of Distress Vocalization

Abstract. *Distress vocalizations were produced in adult guinea pigs by electrical stimulation of the dorsomedial thalamus or the septum-preoptic area. Both of these forebrain-derived vocalizations were increased by systemic administration of naloxone and were inhibited by analgesic periventricular gray stimulation. Naloxone blocked the inhibitory effects of the analgesic stimulation on thalamic vocalizations. Stimulation of nonanalgesic mesencephalic sites in close proximity to the periventricular gray increased the anterior-elicited vocalizations. These data provide evidence for ascending endorphin-mediated inhibition of excitatory forebrain sites for distress vocalizations.*

Numerous studies have confirmed that electrical stimulation of the periventricular gray (PVG) produces analgesia in a variety of species (*1*). This effect seems to be related to activation of endorphin-mediated systems, since such analgesia is at least partially antagonized by naloxone (*2, 3*). Indeed, analgesic PVG stimulation increases the concentration of endorphins in human cerebrospinal fluid (*3, 4*). We now provide evidence that endorphins inhibit brain-stimulated distress vocalizations as well as pain.

We have previously determined that distress vocalizations elicited by social isolation are under endorphin inhibition (*5*). For example, naloxone increases the frequency of such vocalizations in both young guinea pigs and chicks tested in social isolation. It seemed reasonable that the vocalizations (once localized in the brain) would also be responsive to more direct types of endorphin stimulation. In an earlier study (*6*) we used electrical stimulation of the brain to map the adult guinea pig brain for distress vocalization loci. The most reliable areas for obtaining the vocalizations included the ventral septum-preoptic area and the dorsomedial thalamus. Less reliable sites included the dorsomedial hypothalamus, medial forebrain bundle, and certain amygdaloid nuclei. No loci were detected in the cortex or cerebellum. These vocalization sites correspond to those yielding emotional vocalizations in other species (*7*), and correspond to brain areas containing a moderate to high density of opiate receptors (*8*).

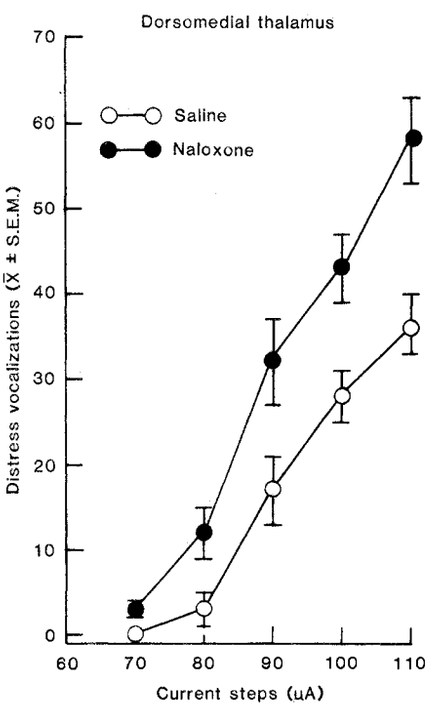


Fig. 1. Effects of saline and naloxone HCl (1 mg per kilogram of body weight) on the frequency of distress vocalizations in adult guinea pigs receiving electrical stimulation of the dorsomedial thalamus. Data were obtained from screening 13 electrode sites in seven animals.

On the basis of these preliminary findings, we analyzed the effects of naloxone and PVG stimulation on distress vocalizations elicited from the septum-preoptic area and the dorsomedial thalamus in adult albino guinea pigs. Animals were surgically prepared with indwelling monopolar electrodes (1-mm tip exposure, skull ground) aimed for the septum-preoptic area or dorsomedial thalamus, and an additional set in the PVG (*9*). One week after surgery, animals were screened for forebrain vocalizations through the use of 60-Hz sine-wave current administered in an ascending 10-$\mu$A current series. During each trial, current was applied during a 30-second period in which six 0.5-second stimulations were administered every 5 seconds. Animals distress vocalized immediately after the end of forebrain stimulation during the 4.5-second intertrial intervals. By comparison, "painlike" screams induced by stimulating mesencephalic sites immediately surrounding the PVG occurred in concert with the stimulation. The pitch of these painlike screams seemed to be higher than that of the distress vocalizations, and resembled vocalizations that accompany the application of an acute nociceptive stimulus such as a clip to the limbs of a guinea pig. Distress vocalizations were also recorded during a 30-second period after stimulation, reflecting the long decay of the response. These poststimulation distress


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vocalizations are summarized in this report, since they were most sensitive to analgesic PVG stimulation. Results of the other response measure have been presented elsewhere (*6*). Trials were separated by 30 seconds, during which the animal did not vocalize. For both forebrain sites, and for both measures, the frequency of the vocalizations increased monotonically as a function of current.

The PVG electrode sites were screened for analgesia according to stimulation techniques discussed above. Analgesia was assessed by a vocalization clip test, in which a clip exerting a pressure of 60 g/mm$^2$ was placed on each of the animal's limbs, and the frequency of clip-induced screams for eight trials was recorded. In this task, control subjects screamed on all eight trials, whereas 10 mg/kg of morphine sulfate (injected subcutaneously) was required to produce a complete analgesia (no screams during eight trials). In separate tests, we evaluated the degree and duration of analgesia produced by PVG stimulation (usually 200 $\mu$A administered for two to ten trials). Complete analgesia lasting between 2 and 15 minutes after PVG stimulation was produced in eight septum-preoptic animals and eight thalamic animals (analgesic groups). Six additional animals in each of the two forebrain groups had PVG electrodes that produced pain-like screams and severe agitation, but no analgesia. These nonanalgesic animals acted as controls and underwent tests identical to the analgesic animals. Subsequent histological analysis revealed that 13 of the 16 mesencephalic electrodes in the analgesic group were within the PVG, whereas 8 of the 12 nonanalgesic sites immediately surrounded the PVG. In all cases, saline or naloxone was administered subcutaneously in the nape of the neck 30 minutes before forebrain stimulation. Data were treated by analyses of variance with factors for both drug and current. Two-tailed *t*-tests were used to evaluate significant ($P < .05$) interactions when a priori unidirectional prediction had been made.

Forebrain-derived distress vocalizations resembled separation distress vocalizations elicited from infant animals (film analysis by five independent, experienced observers). Stimulation of these forebrain sites yielded no abnormal motor responses and seemed to inhibit locomotion. Animals exhibited a head-orienting response (air nosing) to below-threshold currents, which is also observed in infant guinea pigs vocalizing in response to social isolation. By comparison, stimulation of nonanalgesic mesencephalic sites was always accompanied by vigorous jumping and escape attempts.

Distress vocalizations increased as a function of increasing current (Fig. 1). Naloxone induced an overall 75 percent increase in thalamic vocalizations [$\bar{X} \pm$ standard error of the mean: saline, $84 \pm 10$; naloxone, $147 \pm 15$; $F(1, 12) = 26.64$, $P < .001$], and these increases were larger at higher current levels. Thresholds (as defined by the lowest current yielding ten or more vocalizations) were decreased by naloxone ($88 \pm 5$ $\mu$A) as compared with saline ($98 \pm 5$) [$t(12) = 2.52$, $P < .05$]. Naloxone also increased septal-preoptic vocalizations (Fig. 2), although threshold curves for this group were not constructed (*10*).

Figure 2 summarizes the effects of naloxone and analgesic and nonanalgesic PVG stimulation on distress vocalizations as a function of stimulating current (*11*). Naloxone increased the frequency of both thalamic and septal-preoptic vocalizations, and in each case, analgesic PVG stimulation decreased the frequency of vocalizations elicited by brain stimulation. Naloxone blocked the effects of analgesic PVG stimulation on thalamic vocalizations, but did not significantly antagonize the effects of such stimulation on septal-preoptic vocalizations (*12*). In all cases, more statistically powerful effects were obtained when high rather than low current was used to elicit vocalizations. Nonanalgesic PVG stimulation did not increase distress vocalizations significantly during the poststimulation period but did increase them during the stimulation period by 85 percent for septal-preoptic vocalizations [$t(5) = 2.96$, $P < .05$] and by 47 percent for thalamic vocalizations [$t(5) = 3.51$, $P < .02$]. In addition, data obtained during stimulation suggested that naloxone treatment additively increased the potentiating effects of nonanalgesic stimulation on forebrain vocalizations.

Since naloxone increases the frequency of distress vocalizations elicited by electrically stimulating the dorsomedial thalamus or the septum-preoptic area, these vocalizations are probably under endorphinergic inhibition. Further, the reversal of the inhibitory effects of analgesic PVG stimulation on thalamic vocalizations by naloxone is evidence for PVG endorphinergic modulation of this circuit. Analgesic PVG stimulation also decreased septal-preoptic vocalizations, but since naloxone did not block this ef-

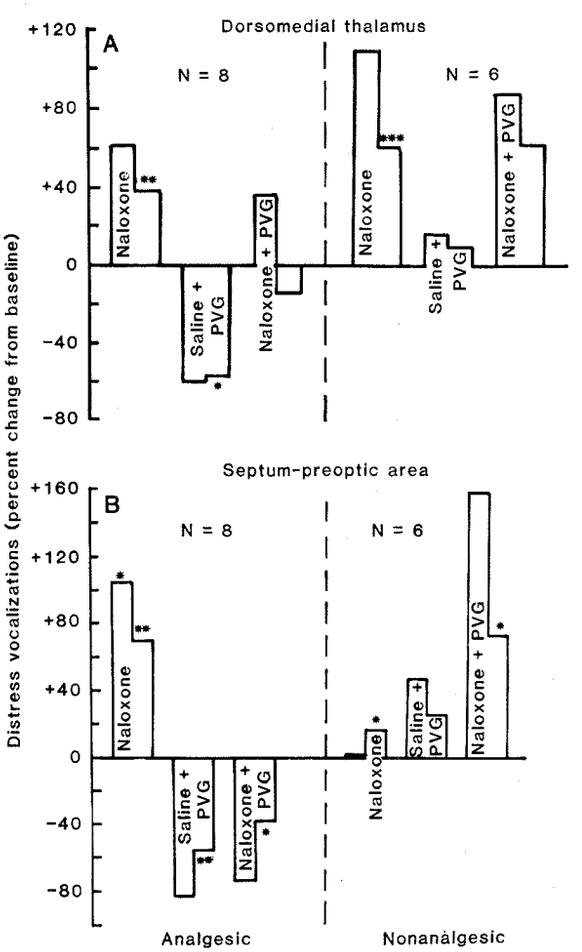


Fig. 2. Effects of naloxone HCl (1 mg/kg) and analgesic and nonanalgesic PVG stimulation on thalamic (A) and septal-preoptic (B) distress vocalizations elicited from adult guinea pigs by electrical stimulation of the brain. The first bar of each pair represents the response to three low-current trials, and the second to three high-current trials. Animals were tested under four counterbalanced conditions: (i) saline plus forebrain stimulation (no PVG stimulation) (*11*), (ii) naloxone plus forebrain stimulation (no PVG stimulation), (iii) saline plus PVG stimulation plus forebrain stimulation, and (iv) naloxone plus PVG stimulation plus forebrain stimulation. Asterisks indicate *P* values associated with two-tailed *t*-tests conducted in comparison with the saline baseline: ***$P < .001$, **$P < .01$, *$P < .05$.

fect, no evidence was provided for PVG endorphinergic involvement in this second circuit. It is not clear what neurotransmitter mediates PVG inhibition of septal-preoptic vocalizations, but serotonin is a likely candidate. For example, there is evidence for serotonin mediation of PVG analgesia (*13*), and administering a serotonin agonist (quipazine) dramatically decreases forebrain vocalizations in guinea pigs (*6*, *14*). Our data extend the observations of others indicating that PVG analgesia inhibits thalamic vocalizations as well as pain by stimulating some ascending endorphinergic system or systems and suggest that the neurotransmitters mediating some of the effects of PVG stimulation depend on the circuitry leading to the final forebrain sites of interaction.

We also found that stimulation of nonanalgesic sites immediately surrounding the PVG produced painlike screams and potentiated the frequency of thalamic and septal-preoptic distress vocalizations. Such stimulation may release some unidentified "distress neurotransmitter" that interacts with forebrain vocalization circuits. Aversive and analgesic loci are close to each other in this region of the mesencephalon, and these sites appear to modulate distant forebrain circuits.

Our results suggest brain circuits controlling distress vocalization and indicate that these circuits are under endorphinergic control. These pathways may participate in the modulation of social distress.

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9. Surgical coordinates were derived from T. J. Luparello [*Stereotaxic Atlas of the Forebrain of the Guinea Pig* (Williams & Wilkins, Baltimore, 1967)]. Septum-preoptic area: anterior (A), 11.8; lateral (L), ± 2.2; ventral (V), 9.0. Dorsomedial thalamus: A, 9.0; L, ± 0.8; V, 6.8. Anterior PVG: A, 5.0; L, ± 0.5; V, 7.5. Posterior PVG: A, 3.0; L, ± 0.5, V, 7.0. Results are presented only for histologically verified placements.
10. As predicted, morphine decreased electrically stimulated distress vocalizations (*6*).
11. Separate saline control sessions were run for each of the manipulations depicted in Fig. 2. For trials with naloxone alone, a single saline session was run in a counterbalanced fashion. In all other cases, saline sessions surrounded each manipulation, and these values were averaged.
12. It is possible that a simple additive interaction between naloxone and PVG stimulation could explain data obtained for the analgesic thalamic animals (Fig. 2). We believe that such additivity is unlikely, however. For example, no evidence for naloxone blockade of analgesic PVG stimulation was obtained in the septum-preoptic group (Fig. 2), despite the fact that naloxone alone increases were larger than PVG decreases in vocalizations.
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## Saliva as a Chemical Cue in the Development of Social Behavior

Abstract. *Throughout development, Mongolian gerbils engage in conspicuous naso-oral investigations of their social partners' mouth areas. The behavioral contribution of saliva-related stimuli in regulating oral-directed responses was studied during several important phases of the gerbil's social life. Weanlings were preferentially attracted to their mother's saliva, subadults at puberty preferred saliva of littermates to that of nonlittermates, and sexually experienced males preferred the saliva of estrous females to that of nonestrous females. The use of saliva as a discriminative cue during various developmental periods suggests that oral chemostimuli have a perennial role in regulating social interchanges.*

Major questions in developmental psychobiology include how social relationships are established and how much adult social behavior is influenced by early social experience. Adequate tests of theories and hypotheses related to these questions require a relatively complete profile of the behavioral processes and mechanisms underlying social interactions at several points during ontogeny and adulthood. We now provide evidence from Mongolian gerbils (social cricetid rodents) suggesting that a chemical communication mechanism mediated by saliva may help to establish and link social behaviors at three stages of the gerbil's life: filial interactions at weaning, sibling or peer interactions at puberty, and sociosexual behavior during adulthood.

Table 1. Investigation responses of sexually experienced male gerbils ($N = 7$) to anesthetized female targets. Abbreviation: AMN, atropine methyl nitrate.

| Target condition | Mean duration of contacts to female body regions (seconds) | | |
|---|---|---|---|
| | Face | Ventral gland | Anogenital region |
| *Experiment 1 (10-minute test)* | | | |
| Estrus | *42.8 | *22.1 | 24.4 |
| Nonestrus | 29.6 | 4.6 | 23.8 |
| *Experiment 2 (5-minute test)* | | | |
| Estrus + AMN | 14.0 | *12.2 | 18.6 |
| Nonestrus + AMN | 13.7 | 0.6 | 16.7 |

*Two-tailed $t$-test, $P < .05$.

While studying social interactions of developing gerbils living in nuclear family units, we recorded a mother-young interchange at weaning (21 to 29 days after birth). During nursing sessions, the pups often engaged in long bouts of licking and nuzzling of the mother's lower lip and chin and lateral mouth region. This filial interchange, also seen in other mammals (*1*), is termed "mouthing" or "mouth-licking." Bouts of mouthing (sometimes lasting 10 minutes or longer) peak during the fourth week after birth as maternal behavior wanes and pups increase their ingestion of solid foods; these bouts are primarily directed to the dam, rarely to the father (*2*).

To examine the relative attractiveness of the dam's facial area during the weaning period, we measured the preferences of 25- or 26-day-old pups to their par-


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