visual cues. In a procedure that separated distance cues and end location cues, both amnesic patients and control subjects accurately reproduced movements in a no-delay condition, but the amnesic patients were impaired relative to controls after delays of 12 and 60 seconds.
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23. S. Corkin (*4*) referred to this phenomenon as "sparing of testing habits" after M. Glickstein and R. W. Sperry [*J. Comp. Physiol. Psychol.* **53**, 322 (1960)].
24. It is not yet clear how the performance of amnesic patients on numerical rule learning or classical conditioning compares with the performance of control subjects. Only when amnesic patients perform as well as controls is it possible to argue conclusively for preserved learning or memory in amnesia. Good, but not normal, performance by amnesic patients could occur because some methods of testing yield good performance in all subjects [L. R. Squire, *Neuropsychologia* **18**, 369 (1980)] and not because the aspect of memory under study is preserved in amnesia. Thus, reports of good performance by amnesic patients on incomplete figures [E. Warrington and L. Weiskrantz, *Nature (London)* **217**, 972 (1968)] or partial information [*ibid.* **228**, 628 (1970); *Neuropsychologia* **12**, 419 (1974)] do not necessarily mean that these tasks demonstrate preserved function. Amnesic patients often do rather well in recognition memory tasks compared with free recall tasks, but the advantage of recognition memory over free recall applies to control subjects as well [G. Talland, *Deranged Memory* (Academic Press, New York, 1965), p. 231].
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Mediation of Diurnal Fluctuations in Pain Sensitivity in the Rat by Food Intake Patterns: Reversal by Naloxone

Abstract. *Rats maintained on a 12-hour light-dark cycle were tested for pain sensitivity after being deprived of food during either the dark or the light phase of the cycle. Diurnal fluctuations in pain sensitivity were observed. The fluctuations followed food intake patterns rather than a natural circadian rhythm, with food deprivation producing a decrease in pain sensitivity. The analgesic response produced by this mild food deprivation was strongly attenuated by naloxone or feeding, suggesting that endogenous opioid systems may be related to patterns of food intake.*

Diurnal variations in pain sensitivity have been demonstrated repeatedly in rats (*1, 2*). These variations are attenuated or abolished by the opiate antagonist naloxone, suggesting that the phenomenon is mediated by an endogenous opioid system (*3, 4*). Strong support for this suggestion is provided by the finding that fluctuations in pain sensitivity are closely paralleled by alterations in levels of endogenous opioids in the rat brain (*5*). It has been assumed that this diurnal cyclicity in pain sensitivity reflects the circadian rhythm common to other pituitary hormone concentrations (*1*). We now present evidence suggesting that diurnal fluctuations in pain sensitivity reflect patterns of food intake rather than a circadian rhythm.

Researchers reporting diurnal rhythms in pain sensitivity and endogenous opioid concentrations used standard light-dark cycles in conjunction with unrestricted feeding (*1, 2*). However, since rats given this liberty feed predominately at night (*6*), the possibility exists that pain sensitivity cycles are entrained by food intake patterns rather than light-dark cycles. This suggestion is supported by the finding that food deprivation can induce analgesia, which is markedly attenuated by naloxone (*4*). Related to this point are several studies implicating endogenous opioid systems in the modulation of food intake. Central injections of opioid peptides increase food intake in the rat (*7*), while opiate antagonists decrease food intake (*8*). Also, the cyclicity in adrenal responsiveness to adrenocorticotropic hormone (ACTH) is primarily a function of food intake patterns (*9*). In view of these findings, we explored the hypothesis that diurnal fluctuations in pain sensitivity are entrained by circadian patterns of food intake.

The subjects were 24 adult male hooded rats individually housed and maintained on a 12-hour light-dark cycle. They were randomly assigned to one of two experimental groups. In the first group (group A), free access to food was given only during the light phase of the cycle; in the second group (group B), only during the dark phase (*10*). Water was always freely available.

After an 8-day period of adaptation to the restricted feeding schedule, the animals were tested for pain sensitivity by the tail-flick method. The latency of tail flicking in response to a thermal stimulus of fixed intensity was determined for each rat during the first 2 to 4 hours of both the light and dark phases (*11*). The food availability schedules for the two groups were then reversed, and the animals were tested again 8 days later.

The results of this experiment clearly indicate that food availability can significantly alter pain sensitivity and that this effect is largely independent of the light-dark cycle. Pain thresholds were uniformly elevated after food deprivation, regardless of the phase of the cycle (Fig. 1A). A within-subjects, repeated-mea-

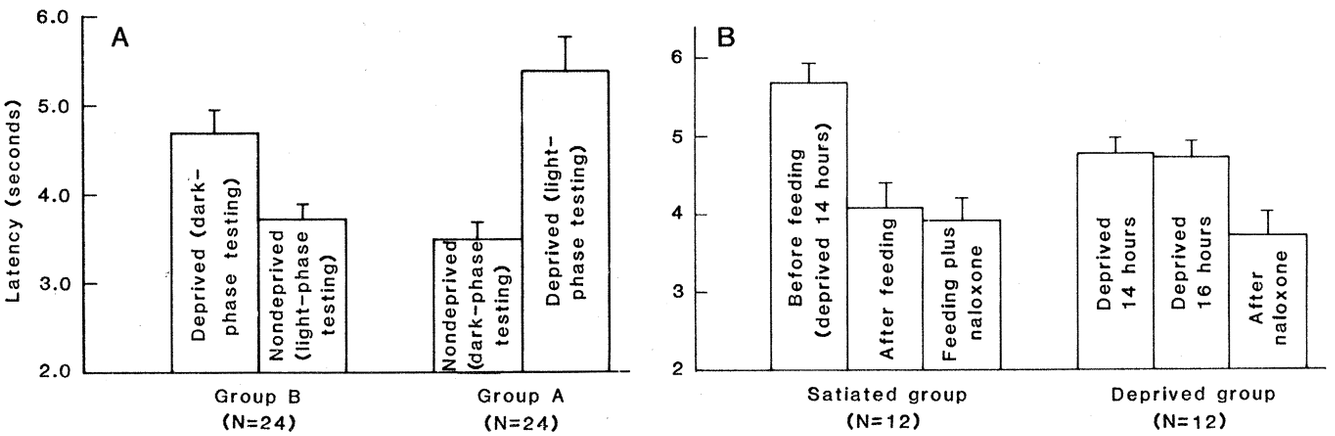

Fig. 1. (A) Effects of light-phase or dark-phase food availability on tail-flick latency in food-deprived and nondeprived rats. Verticle bars represent the standard errors of the mean. (B) Effects of feeding and naloxone administration on tail-flick latency. Since the results for both light phases are comparable, the data are collapsed for illustration. Vertical bars represent the standard errors of the mean.

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sures analysis of variance revealed a significant main effect of food deprivation, $F(1, 23) = 71.87$, $P < .01$. Since rats feed predominantly in the dark when given unrestricted access to food, the animals in group B should have shown the normal cyclicity in pain sensitivity previously reported (*1, 2*). This was in fact the case (Fig. 1A). The lowest pain sensitivity in group B rats was seen when they were tested during the dark phase, $t(10) = 5.87$, $P < .001$, paralleling the reported diurnal elevation in opioid levels (*5*). However, the complete reversal of this pattern in the group A rats reveals the highly significant effect of food intake on this diurnal cyclicity (*12*).

To further evaluate the contribution of hunger to the elevation in pain thresholds, we maintained the animals on the same food schedule for an additional 8 days after completing the testing outlined above. At this point, each group was tested during the 14th to 16th hours of its respective food deprivation period ($N = 12$ per group). The animals were then returned to their home cages, and half of each group ($N = 6$) was allowed access for 2 hours to laboratory chow and a palatable mixture of chocolate milk and mash. (The other half of each group remained deprived of food.) At the end of the 2 hours, all the animals were retested for pain sensitivity. Subsequently, each animal was given an injection of naloxone (4 mg/kg) and retested 15 minutes later. This procedure provided an indication of whether any decrease in pain sensitivity was mediated by an endogenous opioid system.

As shown in Fig. 1B, the rats that were given access to food for 2 hours exhibited a significant increase in pain sensitivity on retesting. Paired t-tests indicate that the decrease in tail-flick latencies after eating was significant for group B, $t(5) = 4.65$, $P < .01$, and for group A, $t(5) = 6.26$, $P < .01$. In contrast, the animals that were not fed showed no significant differences in latency over the 2-hour intertest interval. These findings indicate that food deprivation during either the light or the dark phase results in a decrease in pain sensitivity that can be reversed by food intake.

In the food-deprived animal, naloxone, like feeding, increased pain sensitivity, but it had no effect if pain sensitivity had already been reduced by feeding (Fig. 1B). Both groups that were deprived of access to food during the intertest interval exhibited significant reductions in tail-flick latency after the injection of naloxone (for group B, $t(5) = 3.16$, $P < .02$; for group A, $t(5) = 3.35$, $P < .02$). Naloxone did not significantly reduce latencies in satiated animals tested during either phase of the light cycle.

Our results suggest that daily fluctuations in pain sensitivity to thermal stimuli are strongly mediated by food intake patterns rather than by an independent circadian rhythm. Previous investigations of fluctuations in pain sensitivity or opioid levels in the rat employed similar light cycles in conjunction with unrestricted food availability. Since rats are primarily nocturnal feeders under these conditions, the results from the previous investigations were obtained under conditions functionally paralleling those to which our group B rats were subjected. Thus sensitivity to heat was found to be high during the light phase and low during the dark phase. By restricting the availability of food to the light phase, we were able to reverse the diurnal pattern of pain sensitivity without altering the light-dark cycle.

The increase in pain sensitivity produced by naloxone in food-deprived animals replicates our earlier finding that deprivation-induced analgesia is mediated by an endogenous opioid system (*4*). The fact that feeding also increases pain sensitivity in the deprived animal suggests that the effect of food deprivation on pain sensitivity is related to a hunger state rather than to associated phenomena, such as altered activity cycles. It is not clear whether analgesia induced by food deprivation reflects a generalized stress reaction or is related to a functionally specific role of endogenous opioid systems in food intake. However, β-endorphin and a Met-enkephalin analog produce an increase in food intake after central administration in the rat (*7*). Conversely, naloxone decreases food and water intake in this species (*8*). Recently, Rossier *et al.* (*13*) reported that the concentration of Leu-enkephalin in the pars nervosa of obese adult mice was 200 percent higher than that in littermate controls. Moreover, this increase exhibited a linear trend, with the greatest weight gain occurring in the obese animals. In view of these considerations, it seems likely that the endogenous opioids play a relatively specific stimulatory role in feeding.

Corticosterone levels and cycles of adrenal responsiveness to ACTH are also reported to reflect food intake patterns (*9*). Since ACTH and β-endorphin have the same precursor molecule, reside in the same cells and granules of the pituitary, and are released in equimolar amounts after stress (*14*), it is feasible that this common family of peptides may participate in a complex neurohumoral system that influences food intake and metabolic homeostasis. In this context, it is noteworthy that β-endorphin possesses lipolytic properties (*15*) and has direct stimulatory properties on the pancreas and kidneys (*16*).

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10. Due to the nature of the design, food availability did not correspond exactly to the cycle. To avoid confounding food intake with testing, the phase during which food was available was switched after testing.
11. In tail-flick testing, the latency of the tail's withdrawal from a radiant thermal stimulus of fixed intensity is measured. During testing, the animals were confined in a 3-inch-diameter plastic tube with holes at each end for the head and tail. The animals were given four 15-minute habituation sessions in the restraining tubes before the tests. Each test session consisted of five tail-flick trials, each trial at 2-minute intervals. the five latency values for each animal in each session were then averaged for the analyses.
12. The analysis of variance also revealed an additional small but significant effect of time of testing [light phase–dark phase, $F(1, 23) = 7.32$, $P < .02$]; food deprivation during the dark phase was more effective in elevating tail-flick latencies than deprivation in the light phase. As seen in Fig. 1A, however, this effect is contrary to the normal diurnal cyclicity and was probably due to the fact that the animals were not completely acclimated to the food deprivation schedules. Since the normal pattern of eating is nocturnal (*6*), food availability during the light phase is an unnatural pattern that probably results in greater net deprivation than when food is available during the dark phase.
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