Some ON-OFF receptive fields that could be homologous to those found here have been reported in the LGN of kittens 1 to 2 weeks old (*22*). Failure of such cells to eliminate conflicting inputs could account for the results reported here. Models in which impulse activity brings about selective elimination or inactivation of synapses have been proposed (*2, 23*); elimination of initially excessive inputs has been suggested as a fairly general property of the developing nervous system (*24*).

Translaminar sprouting of remaining optic tract afferents occurs in the LGN of cats unilaterally enucleated during the first week of life (*25*). Action potential blockade may produce a functional enucleation that brings about the same effect, thus accounting for the binocular cells reported here. A different explanation for the binocular cells is suggested by the fact that adult LGN relay cells normally have dendrites that cross laminar boundaries (*26*), which are thought to receive only inhibitory input. Excitatory synapses onto these dendritic regions may be caused or preserved by the action potential blockade.

Action potential activity may contribute to specifying synaptic connections in a variety of ways. For example, blockade could reduce the availability of specific cell-cell recognition markers stored in synaptic vesicles and normally released by action potentials. Another possibility is that utilizable information exists in the pattern of background activity of postnatal ganglion cells. Neighboring ganglion cells of like receptive field type normally tend to fire together (*27*). In the LGN, competitive mechanisms (*2, 23*) may limit relay cell inputs to those having significant coincident activity. This could bring about segregation of ON and OFF and of X and Y pathways, as well as sharpen the retinotopic map.

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Prolactin and Growth Hormone Release by Morphine in the Rat: Different Receptor Mechanisms

Abstract. *Concentrations of prolactin and growth hormone in the serum of rats were significantly increased by morphine. Dose response studies demonstrated that maximum prolactin release required lower doses of morphine than those needed for the maximum growth hormone response. Selective blockade of μ_1 (high affinity) opiate receptors with the irreversible antagonist naloxazone reduced morphine-induced peak concentrations of prolactin by 80 percent while increasing peak growth hormone levels by 250 percent. These results suggest different receptor mechanisms for the opiate modulation of the two hormones. The μ_1 (high affinity) receptor sites appear to mediate the morphine-induced release of prolactin but not growth hormone.*

Opiates and opioid peptides increase the concentrations of prolactin and growth hormone in the serum of rats (*1*). This effect is specific and can be blocked by the opiate antagonist naloxone (*2*). That this opioid action occurs in the hypothalamus (*3*) is interesting in view of the presence of a number of opioid peptides in this region (*4*). Also present in this region are opiate receptors through which these agents produce their effects (*5*). Recent studies suggest that there are distinct subpopulations of opiate binding sites (*6*) which vary in their pharmacological profiles, regional distribution, ontogeny, phylogeny, and sensitivity to proteolytic enzymes and reagents (*7*). We now present evidence that the morphine-induced release of prolactin and growth hormone are mediated through separate and pharmacologically distinct opiate receptors.

Indwelling jugular cannulas were placed in male Sprague-Dawley rats (180 to 220 g; Charles River Breeding Laboratories) anesthetized with ethane and oxygen. Cannulas were routinely filled with a dilute solution of heparin in saline (25 U/ml) to prevent clotting. After implantation of the cannulas, the animals received either nothing, naloxone (50 mg/kg), or naloxazone (50 mg/kg) intravenously. Naloxazone solutions were made by dissolving free base in saline

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containing 1 percent acetic acid (10 mg/ml) 15 to 20 minutes before injection. All hormone experiments were performed 24 hours after implanting the cannulas to permit the animals to recover from surgery and to ensure the elimination of free, unbound naloxone or naloxazone. Heparinized blood samples (0.5 ml) were obtained, the animals were given morphine sulfate (10 mg/kg, intravenously), and additional blood samples were taken at the specified times (Figs. 1 and 2). Both serum prolactin and growth hormone concentrations were then determined by radioimmunoassay (*8*) in the same serum sample to ensure that possible differences between the prolactin and growth hormone response did not result from variability between animals.

Earlier reports of dose response curves for morphine-induced prolactin and growth hormone release indicated that morphine's ability to increase prolactin was greatest at 10 mg/kg and showed no further increase with a higher dose (15 mg/kg) (*2*). Growth hormone levels, however, increased 50 percent more when the dose of morphine was increased from 10 to 15 mg/kg. Our results confirm these findings (Fig. 1).

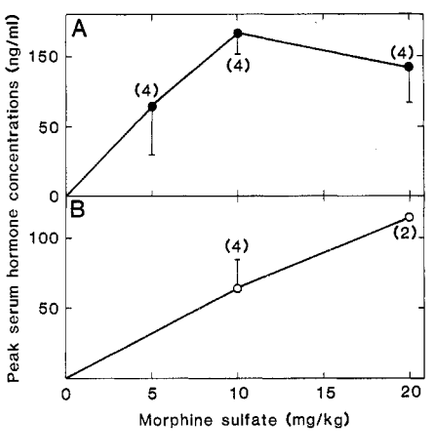

Fig. 1. Dose response of morphine-induced (A) prolactin and (B) growth hormone release. Rats were prepared with indwelling jugular cannulas and treated with the appropriate dose of morphine sulfate the next day. Results are the peak (± standard error of the mean) serum hormone values at 15 to 30 minutes after injection. The number of animals tested at each point is indicated. Baseline prolactin values (mean ± standard error) were 5 mg/kg, 4.8 ± 0.1 ng/ml; 10 mg/kg, 5.5 ± 0.75 ng/ml; 20 mg/kg, 10.8 ± 5.7 ng/ml. The baseline growth hormone value for the group receiving 10 mg/kg was 7.9 + 1.3 ng/ml, and for the group receiving 20 mg/kg, 7.9 mg/ml. Prolactin concentrations after morphine administration were significantly higher than basal levels at the 10 mg/kg ($P < .001$) and 20 mg/kg ($P < .02$) doses but not at the 5 mg/kg ($P < .1$) dose. The growth hormone levels after morphine administration (10 mg/kg) were also significantly higher than resting levels ($P < .02$).

With increasing morphine doses, peak prolactin levels increased, plateauing at 10 mg/kg. Peak growth hormone levels, by contrast, increased almost twofold when the morphine dose was increased from 10 to 20 mg/kg. Although consistent with a number of explanations, these results suggested different sensitivities to morphine for the two hormones, possibly reflecting a higher affinity receptor mechanism for opiate modulation of prolactin release than growth hormone release.

Saturation studies of opiate binding in rat brain have clearly indicated high affinity [dissociation constant (K_D) < 1 n*M*] and lower affinity binding (K_D < 10 n*M*) for a number of opiate and opioid peptide ligands (*6*). Additional evidence suggests that opiates, such as morphine, and enkephalins bind with highest affinity (K_D < 1 n*M*) to a common site, recently termed the μ_1 site, and with slightly lower affinity (K_D < 10 n*M*) to additional sites that preferentially bind either morphine (μ_2) or enkephalin (δ) (*9*). Naloxazone selectively and irreversibly blocks the μ_1 (high affinity) site both in vivo and in vitro (*10*). In previous studies of naloxazone action in rats the investigators injected high doses subcutaneously (*10*). The intravenous administration of naloxazone used in the present studies increased its potency fivefold, eliminated absorption problems, and had pharmacological effects that were identical to those described after subcutaneous injections. At 50 mg/kg, naloxazone eliminated all μ_1 binding, as determined by either Scatchard analysis or competitive displacement curves (*11*). Since previous studies with subcutaneous administration demonstrated a marked loss of morphine's analgesic potency with blockade of μ_1 sites, we tested morphine analgesia in animals treated the day before with either naloxone or naloxazone (50 mg/kg) intravenously (*12*). At both 5 and 10 mg/kg morphine sulfate was analgesic in the tail-flick assay in all rats treated the previous day with naloxone ($N = 7$ and $N = 4$, respectively) and in none of the rats in the naloxazone- ($N = 7$ and $N = 5$; $P < .001$ and $P < .01$, respectively) treated group.

Morphine (10 mg/kg) significantly increased prolactin from its basal value of 27.5 ± 8.1 ng/ml to 152 ± 31.8 ng/ml at 15 minutes and 117 ± 25.7 ng/ml at 30 minutes in the rats treated 24 hours earlier with naloxone (Fig. 2). These concentrations are quite similar to the peak levels in animals given no naloxone but injected with morphine at the same dose (119 ± 12.7 ng/ml). Animals treated with naloxazone also showed a significant increase in prolactin from basal levels of 8.3 ± 2.3 ng/ml to 29.0 ± 8.1 ng/ml at 15 minutes and 20.0 ± 4.2 ng/ml at 30 minutes. However, compared to naloxone-treated controls, naloxazone treatment decreased the peak prolactin level by more than 80 percent. Thus, blockade of μ_1 sites with naloxazone markedly attenuated morphine's ability to increase prolactin.

The effect of naloxazone treatment on morphine-induced growth hormone release in the same animals was quite different from that of prolactin. Instead of the dramatic inhibition of prolactin release, peak growth hormone values in naloxazone-treated animals (648 ± 161 ng/ml) were increased 250 percent relative to the naloxone-treated group (260 ± 89 ng/ml; $P < .10$).

Naloxone treatment alone appeared to have a delayed effect on resting hormone levels long after the elimination of the antagonist from the animal. Comparison of the basal prolactin levels prior to the injection of naloxone (2.8 ± 0.4 ng/ml)

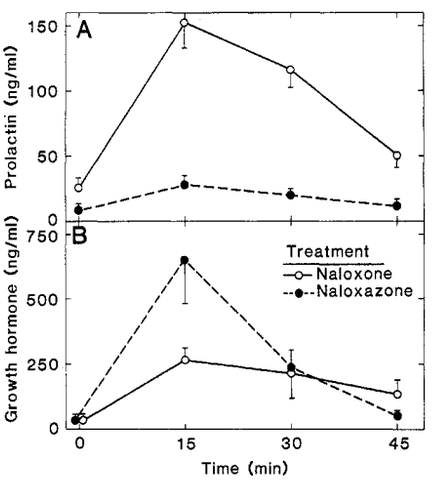

Fig. 2. Effect of naloxone and naloxazone on (A) prolactin and (B) growth hormone release. Groups of rats ($N = 6$) were prepared with indwelling jugular cannulas and were then treated with either naloxone or naloxazone (50 mg/kg). Blood samples were taken 24 hours later for determination of baseline hormone concentrations. The animals were then given morphine sulfate (10 mg/kg intravenously) and additional blood samples were taken 15, 30, and 45 minutes after the morphine administration. Each serum sample was assayed for both prolactin and growth hormone. The peak prolactin levels, seen at 15 minutes, were significantly higher then baseline values in both the naloxone- ($P < .002$) and naloxazone- ($P < .02$) pretreated groups. The peak prolactin values between both groups also differed significantly ($P < .005$). Similarly, the peak growth hormone levels, also seen at 15 minutes, were significantly elevated over baseline values for the naloxone ($P < .02$) and the naloxazone group ($P < .003$). There are no significant differences ($P < .10$) between peak growth hormone levels in the two groups.

and 24 hours later in the same animals (27.5 ± 8.1 ng/ml) demonstrated a ninefold increase ($P < .02$). This effect seemed specific for naloxone. Basal prolactin levels 24 hours after implanting the cannulas in untreated control animals (7.0 ± 1.8 ng/ml; $N = 12$) were not significantly different from the values recorded prior to naloxone administration in the other groups of animals. A threefold elevation in resting growth hormone levels after naloxone administration, from 8.3 ± 3.2 ng/ml to 24.3 ± 8.5 ng/ml ($P < .05$), was also noted. These delayed effects of antagonist administration on resting hormone levels seem to have the same receptor selectivity as the morphine-induced release. Basal prolactin concentrations after naloxone injection were over threefold greater than those after naloxazone injection ($P < .05$), whereas growth hormone levels in the two groups were virtually identical.

Our results imply that the receptor mechanisms for morphine-induced prolactin and growth hormone release are different. The sensitivity of both prolactin release and analgesia to naloxazone suggests their mediation through the μ_1 sites, whereas growth hormone concentrations appear to be modulated through a lower affinity receptor. These conclusions are supported both by our data (Fig. 1) and by the data of others (*1, 2*) demonstrating a maximum elevation of prolactin at lower morphine doses than those required for growth hormone.

Naloxone's delayed action on hormone concentrations raises many questions. It may reflect a physiological rebound following the sudden suppression of hormones by naloxone (*1, 2*). However, these actions might also reflect an increased sensitivity of the system to opioids at the receptor level. Early studies of opiate receptor binding demonstrated dramatic increases in binding associated with the in vivo administration of opiates and particularly with the administration of antagonists (*13*). Perhaps this increase in binding sites is responsible for an increased sensitivity of the system to hormonal release as a result of opioid administration. It has been proposed that both prolactin and growth hormone concentrations are under tonic control by endogenous opioids (*1, 2*). Increased receptor sensitivity, therefore, would be expected to correspond with elevated basal hormone levels. This possibility also might explain why peak growth hormone concentrations after morphine administration are elevated in animals previously treated with an antagonist compared to untreated control animals. Increases in peak morphine-induced prolactin levels after naloxone treatment would not be expected in these studies since the dose of morphine used (10 mg/kg) produced a maximum response in the untreated group (Fig. 1).

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Mice Regrow the Tips of Their Foretoes

Abstract. *Mice will replace the tip of a foretoe when it is amputated distal to the last interphalangeal joint. Amputation of the digit more proximal to the joint does not result in regrowth of the foretoe. Though this growth shares certain similarities with the epimorphic regeneration of amphibian limbs, the two processes are not the same. The regrowth reported here in mice is probably similar to the scattered clinical reports of fingertip regeneration in children, and presents a model system with which to explore the controls of wound healing and tissue reconstruction in mammals.*

It is commonly believed that mammals do not regenerate their extremities in the same way that lower vertebrates or invertebrates do. Among chordates, regenerative ability has reached its zenith in the tailed amphibians. For example, most adult salamanders will regenerate a complete limb within 3 months after amputation. Initially, the limb stump is covered by an epithelium which later stratifies to a thickened apical cap as regeneration proceeds (*1*). However, if skin of full thickness is grafted over the wound this inhibits the regeneration of the limb (*2*). Accidental amputations of human fingers are often treated clinically in just this manner: the amputation surface is closed off from the environment by a sutured skin flap. The result of this surgical manipulation is a stump (*3*). Some clinicians (*3, 4*) have debrided the amputation surface, changed dressings frequently, but performed no surgical intervention. The surprising result, observed in both children and adults, has been regrowth of the fingertip.

In humans, as in metamorphosizing anuran larvae (*5*), the level of amputation

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