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CENTRAL NORADRENERGIC TERMINAL EXCITABILITY: EFFECTS OF PRESYNAPTIC RECEPTOR STIMULATION AND BLOCKADE

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1. INTRODUCTION

Numerous biochemical studies using brain slices preincubated with tritiated norepinephrine (NE) have assessed the effects of various drugs, including adrenergic drugs, upon the stimulation-evoked release of NE from the nerve endings of central noradrenergic neurons (Langer, 1977; Westfall, 1977; Starke, 1977). In this in vitro preparation, NE itself and alpha$_2$-adrenoceptor agonists reduce the electrical stimulation or potassium-induced outflow of tritium, whereas alpha$_2$ antagonists enhance the stimulus-evoked overflow of tritium (Farnebo and Hamberger, 1971; Starke and Montel, 1976; Dismukes and Mulder, 1976; Taube et al., 1977; 1977; De Langen et al., 1979; Mulder et al., 1979; Shenoy and Ziance, 1979; De Langen and Mulder, 1980). Based on these findings in vitro it was proved that receptors located on presynaptic nerve terminals that are sensitive to a neuron's own neurotransmitter might exist (i.e. autoreceptors).

In addition to adrenergic drugs, other agents such as opiates and prostaglandins have been shown to exert an analogous effect upon NE release from noradrenergic nerve terminals (Langer, 1974; Montel et al., Montel et al., 1975; Taube et al., 1977; Arbilla and Langer, 1978; Starke, 1980). Thus, presynaptic terminal receptors, which can be stimulated not only by the neuron's own neurotranasmitter but also by other agents, appear to participate in the regulation of NE release in central noradrenergic neurons.

A regulatory role for presynaptic terminal receptors in transmitter release has also been proposed for other central monoaminergic neurons (Farnebo and Hamberger, 1971; Langer, 1977; Starke et al., 1978; Westfall et al., 1982; Gonon and Buda, 1985). Release of dopamine (DA) and serotonin (5-HT) from dopaminergic and serotonergic nerve endings is altered in a similar way to that of NE by dopaminergic and serotonergic autoreceptor agonists and antagonists. Thus, a process analogous to that occurring in the terminals of central noradrenergic neurons is likely to occur in the terminals of dopaminergic and serotonergic neurons following autoreceptor activation (see next chapter).

Despite an accumulation of biochemical evidence for the existence of presynaptic terminal receptors, the mechanisms underlying the modulatory role of terminal receptors in NE release remain unknown. In addition, although the biochemical consequences of the activation of presynaptic terminal receptors have been desmonstrated both in vitro and in vivo (Besson et al., 1978; Chesselet et al., 1982), it is not known whether these receptors normally function physiologically in vivo. Intracellular recording from nerve endings or terminal axons would be an ideal means of investigating the

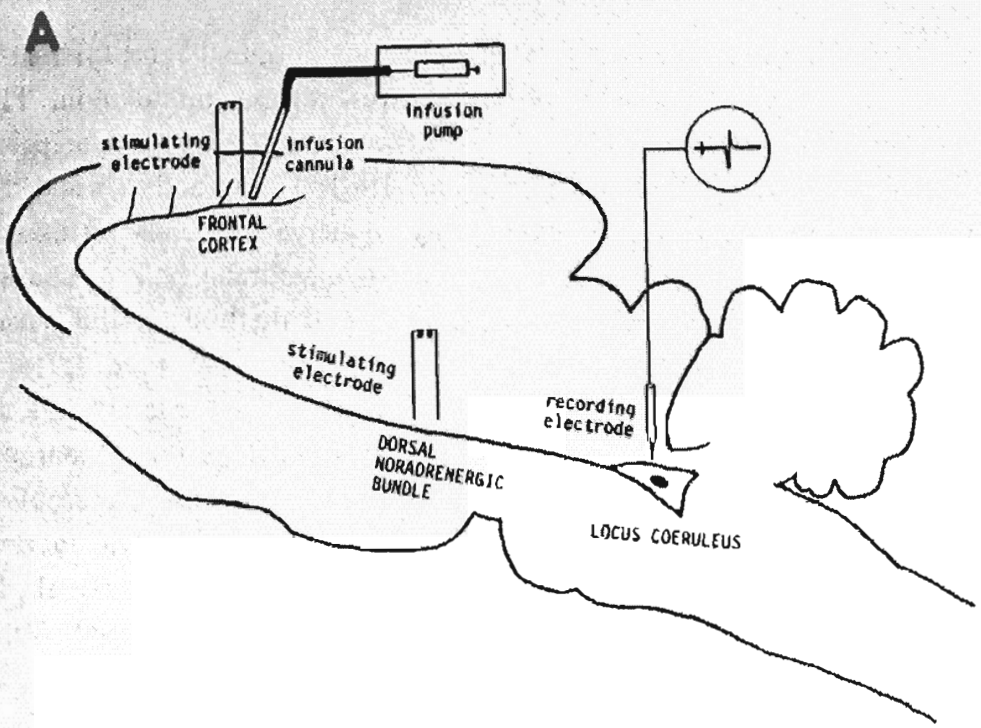
neurophysiological consequences of activating terminal receptors, so as to understand the mechanisms of transmitter release modulation. This is, however, not possible at present except in some invertebrate preparations (Llinas et al., 1976; Baxter and Bittner, 1981; Dixon and Atwood, 1985). Alternatively, changes in the excitability of nerve terminals, resulting from presynaptic receptor-mediated alterations in membrane polarization and/or conductance, can easily be measured. The ideal method for this purpose is one derived from the approach of Wall (1958) who used it to study presynaptic inhibition in the spinal cord. Over the past several years, this in vivo technique has been applied in neuropharmacological and neurophysiological studies of the effects of stimulation and blockade of autoreceptors and other presynaptic receptors on the nerve terminals of central monoamine neurons (Groves et al., 1981; Nakamura et al., 1981; Nakamura et al., 1982a; Nakamura et al., 1982b; Tepper et al., 1984a; Tepper et al. 1984b; Tepper et al., 1985; Ryan et al., 1985a; Ryan et al., 1985b; Sawyer et al., 1985; Tepper et al., 1986).

In this chapter, this useful experimental method will be discussed together with a review of recent results on the neuropharmacology and neurophysiology of the presynaptic receptors of central noradrenergic terminals. This review will focus on those receptors sensitive to adrenergic agents and opiates. In the following chapter the literature on the effects of stimulation and blockade of the terminal autoreceptors on central dopaminergic and serotonergic neurons will be reviewed, followed by a discussion of the possible mechanisms underlying presynaptic receptor-mediated changes in the terminal excitability of central monoamine neurons.

2. MEASUREMENT OF TERMINAL EXCITABILITY

The following description of the technique for the measurement of presynaptic receptor-mediated changes in the terminal excitability of central nervous system neurons is specific for the frontal cortical terminals of noradrenergic neurons, but is applicable to many other systems as well.

The locus coeruleus is the major source of noradrenergic projections in the brain. Since the locus coeruleus projects to the central cortex, the properties of central noradrenergic autoreceptors and other presynaptic receptors are best studied in noradrenergic terminals of the cerebral cortex. Thus, we have studied the electrophysiological effects of auto- and presynaptic receptor stimulation in the frontal cortical fields of noradrenergic neurons of the locus coeruleus. Figure 1 schematically illustrates the procedure.



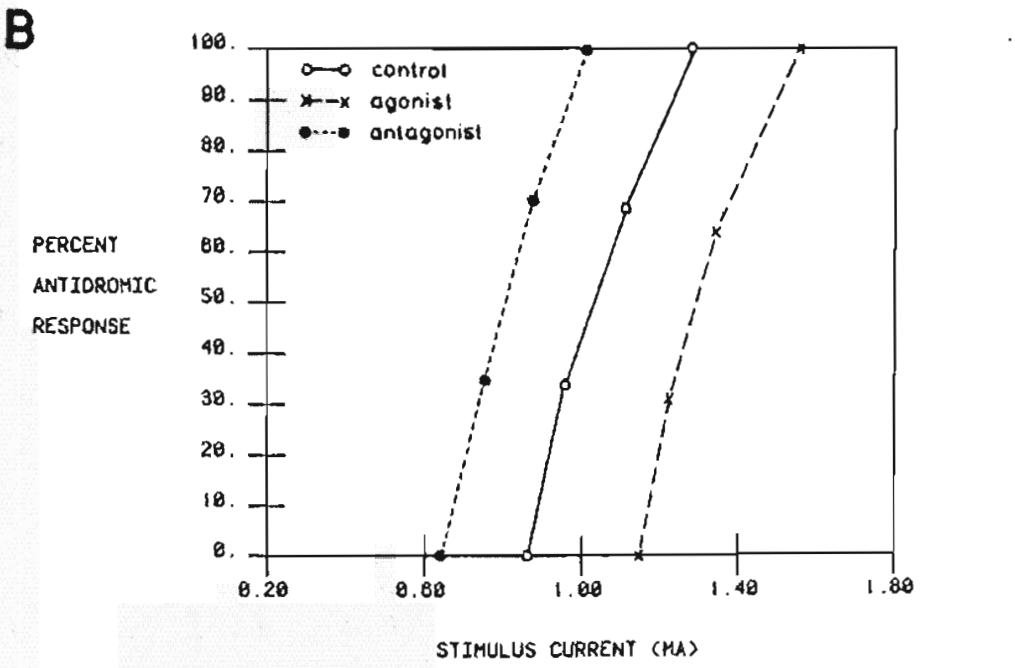


Fig. I. Technique for measuring the terminal excitability of noradrenergic axons in anesthetized rats. A. Schematic diagram of experimental procedure. Single unit activity is extracellularly recorded in the locus coeruleus, and bipolar stimulating electrodes are implanted into the dorsal noradrenergic bundle and into the terminal fields of locus coeruleus neurons in the frontal cortex. Drugs are infused into the frontal cortex through an infusion cannula connected to an infusion pump. B. Hypothetical excitability curves showing decreased terminal excitability produced by an agonist and increased terminal excitability by an antagonist.

A bipolar stainless steel enamel-coated stimulating electrode, with a tip separation of less than 100 μm, was placed into the frontal cortex of urethane-anaesthetized rats, and an extracellular recording electrode was advanced until a single, electrophysiologically identified noradrenergic locus coeruleus neuron was encountered that responded antidromically to frontal cortex stimulation (Nakamura and Iwama, 1975; Nakamura, 1977; Sakaguchi and Nakamura, 1987). When a stimulus of sufficient intensity (0.2–3.0 mA; 0.05-1.0 ms) was delivered to the terminal field, an antidromic action potential was elicited in an all-or-none fashion. However, over many trials (30-200), the number of stimuli effectively eliciting antidromic action potentials will vary as a result of small fluctuations in the excitability at the site of impulse initiation. First, the "threshold" – the minimum current necessary to evoke antidromic responses on 100% of trials was determined (excluding trials on which a collision between a spontaneous spike and the antidromic response occurred). In addition to this value, the proportions of antidromic responses evoked by several lower stimulus currents were calculated. It was then possible to construct a curve showing the relationship between stimulus currents and the proportion of antidromic responses, similar to a dose-response curve. In this format, a decrease in terminal excitability is indicated by an increase in threshold, and a shift to the right in the current-response curve, while increased terminal excitability is characterized by threshold decrease and a leftward shift in the excitability curve (Figure 1B).

Drugs were applied either intravenously or by direct infusion into the frontal cortex at low concentrations (0.1-50 μM) and small volumes (300-600 nl) by means of one or two small cannulae situated approximately 50-100 um from the exposed tips of the bipolar stimulating electrode. Terminal excitability was measured before and after drug application, and the effects of these drugs qualitatively assessed, based on changes in threshold and the excitability curves, as shown in Figure 1B. A quantitative measurement of changes in terminal excitability is obtained by dividing the post drug threshold current by the pre-drug threshold current and is referred to as percent change in terminal excitability.

3. STIMULATION OF TERMINAL AUTORECEPTORS

3.1. Effects of Adrenergic Drugs

3.1.1. Alpha-agonists

When infused directly into the cortex, the alpha$_2$-agonist clonidine (10 μM) reliably caused a decrease in noradrenergic terminal excitability, as shown in Figures 2 and 3A (Nakamura et al., 1981). The decrease in terminal excitability produced by clonidine was often accompanied by a slight prolongation (0.5-1.0 ms) in antidromic latency and by an increase in the variability of the antidromic response latency. The decreased terminal excitability induced by clonidine could be reversed by the alpha-antagonist, phentolamine, as illustrated for a typical locus coeruleus neuron in Figure 4.

It is known that the spontaneous firing rate of locus coeruleus neurons can be suppressed by alpha$_2$-agonists acting on soma-dendritic autoreceptors, and that the agonist's effect can be reversed by appropriate alpha-antagonists (Aghajanian et al., 1977; Cedarbaum and Aghajanian, 1976, 1977). Furthermore, recent intracellular recording studies in vivo and in vitro have desmonstrated that the alpha$_2$-autoreceptor-mediated inhibition of locus coeruleus firing results from a hyperpolarization of the soma-dendritic membrane due to an increase in potassium conductance (Aghajanian and Vandermaelen, 1982; Egan et al., 1983; Williams et al., 1984; 1985) It is possible that a similar mechanism underlies the alterations in NE terminal excitability produced by stimulation of terminal autoreceptors. Decreased terminal excitability, accompanying an increase in antidromic latency and in latency variability, may reflect an autoreceptor-mediated hyperpolarization of the terminal membrane, whereas increased terminal excitability may result from depolarization at the terminal, arising from autoreceptor disinhibition. In consonance with this hypothesis, local infusion of potassium, a potent depolarizing agent was found to increase the excitability of locus coeruleus terminals, as shown in Figure 3D (Nakamura et al., 1981). However, other mechanisms such as a change in conductance could also be involved in autoreceptor and other presynaptic receptor-mediated alterations in terminal excitability. The mechanisms of the changes in terminal excitability and evoked transmitter release induced by presynaptic receptor stimulation and blockade are discussed in greater detail in the next chapter in this volume (Tepper et al., this volume).

When infused by itself, the alpha-antagonist phentolamine produces an increase in noradrenergic terminal excitability, illustrated in Figure 3B

(Nakamura et al., 1981). This increased terminal excitability is often accompanied by a slight decrease in antidromic latency and a decrease in the latency variability. These results suggest that endogenous NE released from locus coeruleus terminals tonically stimulates noradrenergic terminal autoreceptors, resulting in a decrease in terminal excitability to a certain

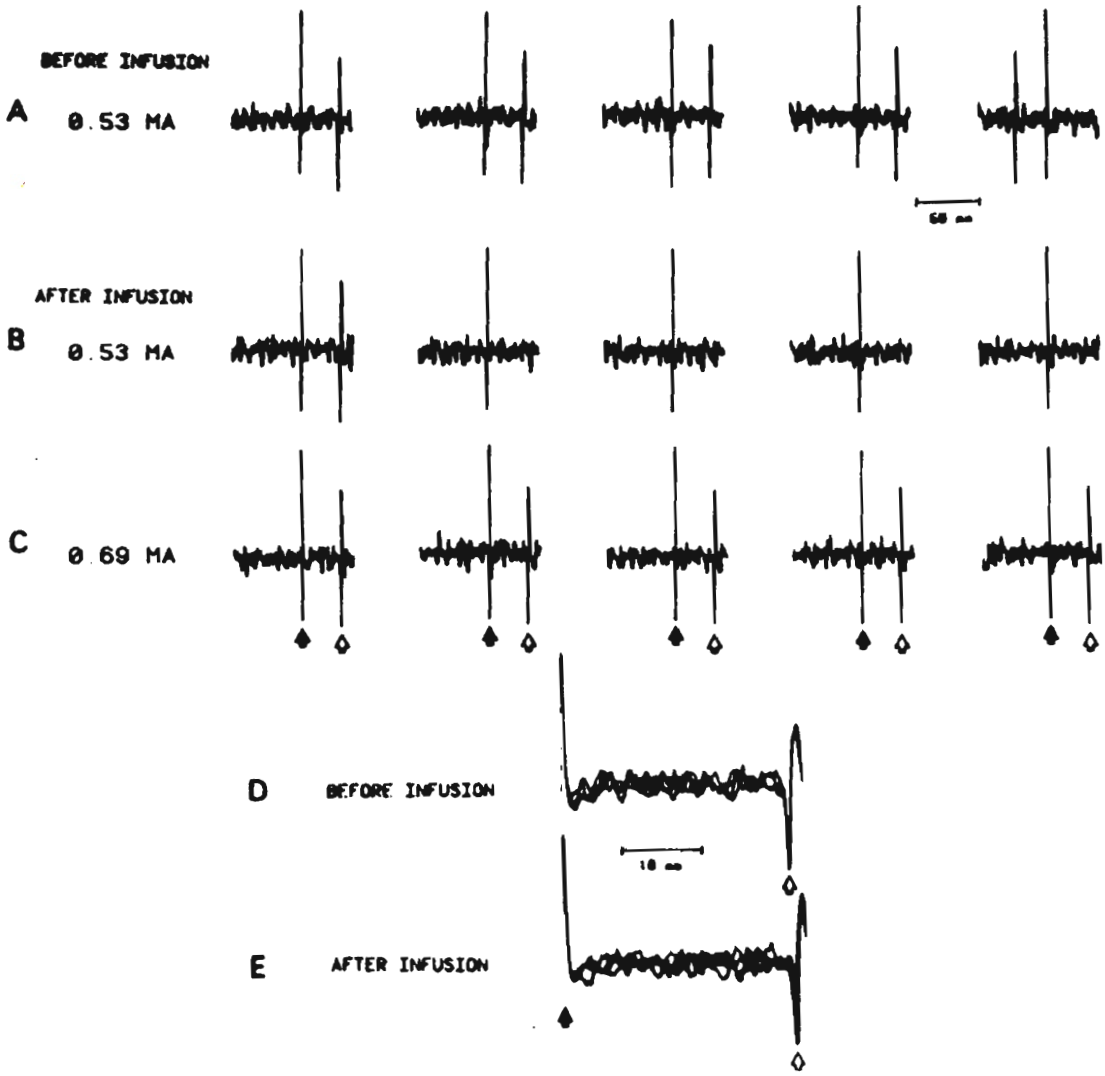


Fig. 2. Antidromic action potentials evoked by stimulation of the terminal field in the frontal cortex are shown before and after local infusion of the alpha$_2$-agonist clonidine (10 μM). Before infusion, the minimum current sufficient to evoke 100% antidromic invasion on non-collision trials was 0.53 mA (A). After clonidine infusion, this current became ineffective (B) and a current of 0.69 mA was required to produce 100% antidromic responding (C). The antidromic latency and the variability of the latency are shown prior to (D) and following (E) clonidine infusion. Black arrows indicate stimulus artifact, and white arrows indicate antidromic action potentials. (From Nakamura et al., 1981, with permission.)

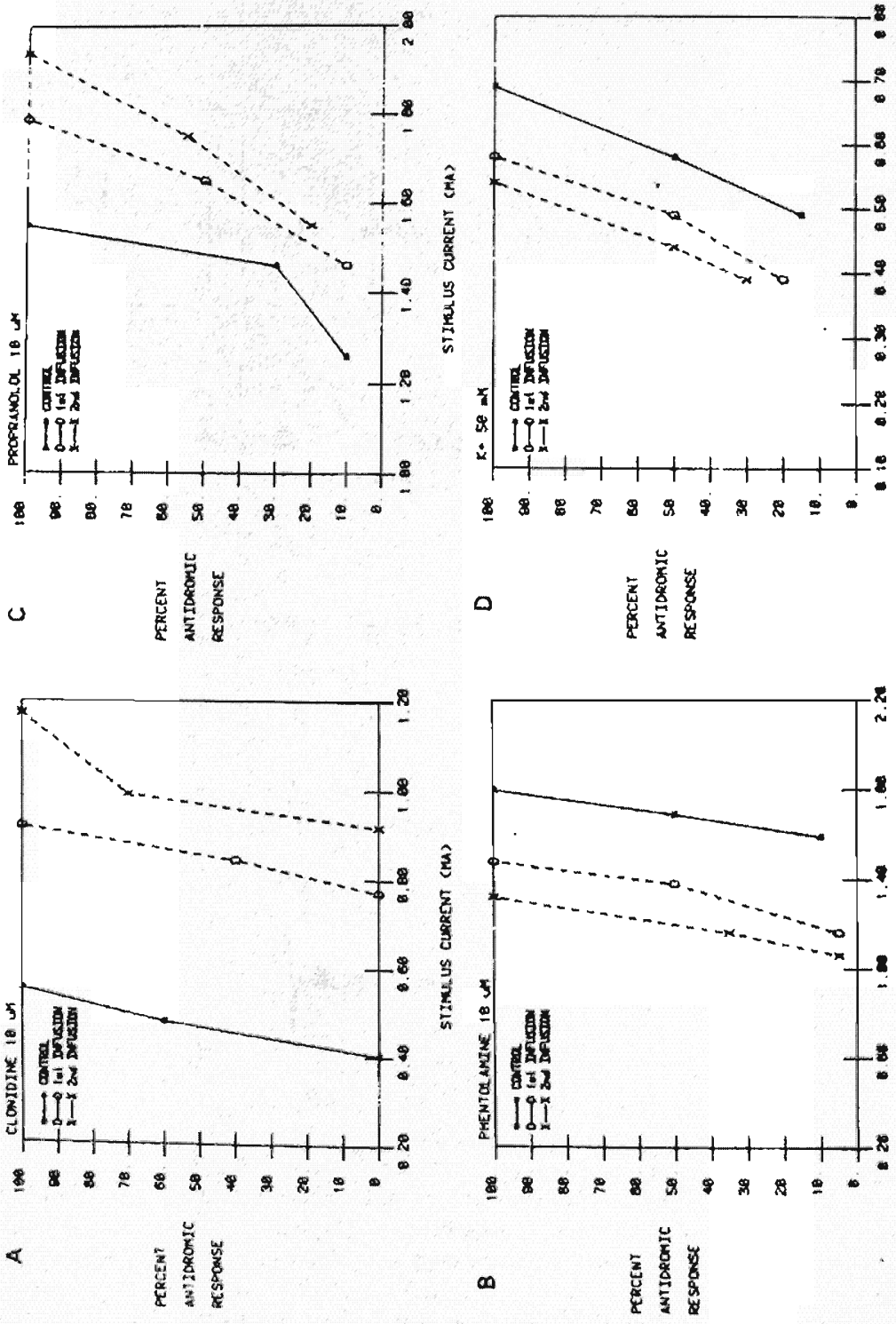


Fig. 3: Typical terminal excitability curves before and following local infusion of various drugs into the frontal cortex. A. Infusion of 10 uM clonidine results in a shift in the curve to the right, indicating decreased terminal excitability. B. The alpha$_2$-antagonist, phentolamine (10 μM) by itself produces increased terminal excitability, as revealed by a parallel shift in the curve to the left. C. Infusion of the alpha-antagonist propranolol (10 μM) causes decreased terminal excitability. D. Local infusion of 50 mM potassium, a potent depolarizing agent, into the terminal field in the frontal cortex produces increased terminal excitability, an effect similar to that

level. This observation is particularly important in providing evidence that the physiological activation of terminal autoreceptors occurs in vivo.

3.1.2. Beta-Agonists

There is also some evidence for the existence of presynaptic beta-receptors on noradrenergic terminals. However, in contrast to the alpha$_2$-autoreceptors, the existence of a presynaptic beta-receptor on noradrenergic terminals is somewhat controversial. Although some studies have shown that stimulation of presynaptic beta-receptors increases NE release under some conditions, this is not always the case (Langer, 1977; Shenoy and Ziance, 1979).

Local infusions of the beta-agonist isoproterenol (0.1-10μM) failed to produce any significant changes in noradrenergic terminal excitability. However, cortical infusions of the mixed beta-antagonist propranolol (1-10 μM) led to reliable and significant decreases in the excitability of locus coeruleus terminals, as shown in Figure 3C (Nakamura et al., 1981). Although this finding is somewhat paradoxical, it is consistent with the in vitro biochemical release studies in that these studies have shown that presynaptic

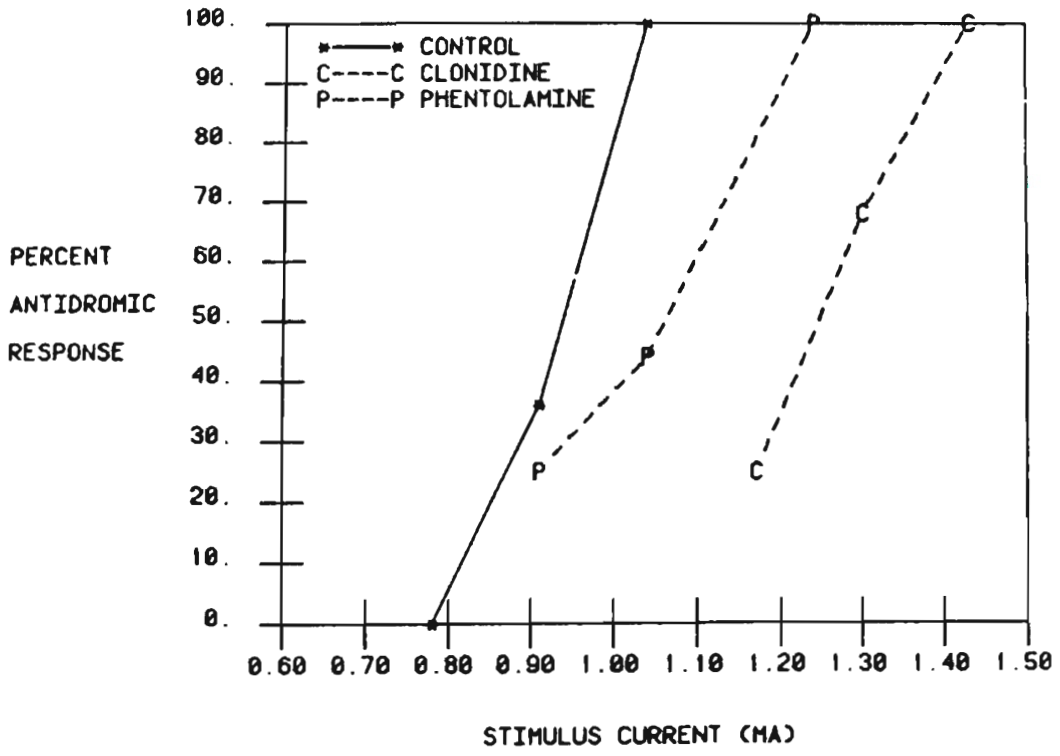
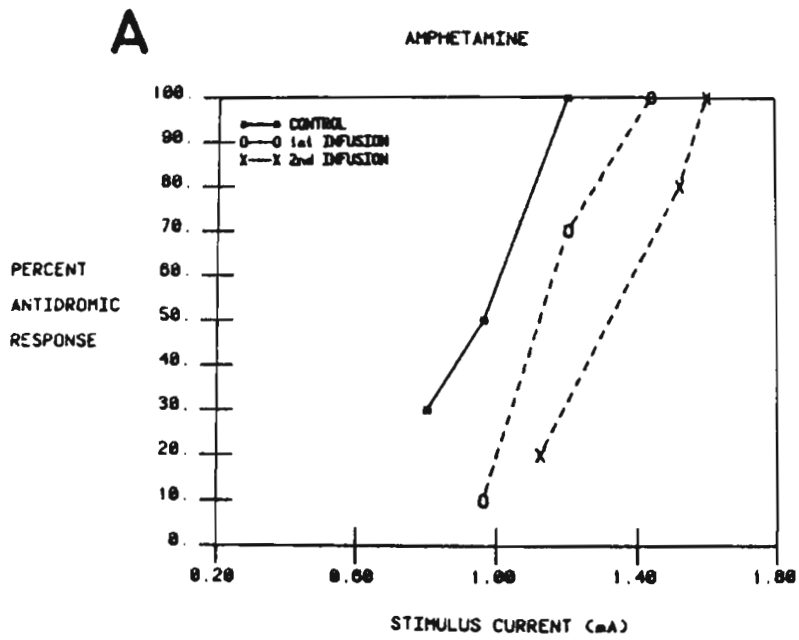


Fig. 4. Antagonistic effects of 10 μM phentolamine upon decreased terminal excitability produced by prior infusion of 10 μM clonidine. (From Nakamura et al., 1981, with permission).

beta-receptor stimulation and blockade lead to exactly the opposite effects on transmitter overflow as do alpha$_2$-autoreceptor stimulation and blockade. From an operational point of view, it has been proposed that the function of the presynaptic beta-receptor is to increase NE release when the extracellular concentration of the transmitter is low, and that the effects of beta-receptor stimulation are only seen when extracellular levels of NE are below the threshold for activating the inhibitory alpha$_2$-autoreceptors. Measurements of postsynaptic alpha- and beta-adrenoceptors indicate that the affinity of NE is some 100 times greater for the beta-receptor than for the alpha-receptor (Langer, 1977). However, under the in vivo conditions of the terminal excitability experiments, there is a sufficient extracellular concentration of NE to stimulate the alpha$_2$-autoreceptor, since application of phentolamine by itself leads to increased terminal excitability. Therefore, it is most likely that the beta-agonist failed to produce any changes in terminal excitability because all the beta-receptor sites were already occupied by endogenously released NE. It is precisely under these conditions, however, that beta-receptor antagonists would be expected to exert their maximal effects, as we observed. Thus, alpha$_2$- and beta-presynaptic receptor stimulation produce opposite effects on noradrenergic terminal excitability, just as they do on evoked release of NE.

3.2. Effects of Amphetamine

Since amphetamine is known to provoke release and prevent re-uptake of catecholamines (Arnold et al., 1977; Carr and Moore, 1969; Chiueh and Moore, 1974; Heikkila et al., 1975; Kuczenski, 1983; Rutledge et al., 1972), we attempted to stimulate noradrenergic terminal autoreceptors by endogenous NE accumulated within the terminal fields after amphetamine administration. Similar to administration of the alpha$_2$-agonist clonidine, the excitability of NE terminals in the frontal cortex was decreased in a dose-related fashion by the local infusion of amphetamine into the terminal fields, as shown in Figure 5 (Nakamura et al., 1982a). Because this effect is prevented by depletion of endogenous NE by pretreatment with alpha-methyl-p-tyrosine (Figure 5B), the decreased terminal excitability produced by amphetamine administration is indirect, and likely results from increased autoreceptor stimulation due to the accumulation of extracellular NE at the terminals. In addition, prior infusion of phentolamine into the terminal fields blocks amphetamine's effect on terminal excitability. Thus, the decreased terminal excitability caused by amphetamine infusion is not due to a direct effect of amphetamine, but due to endogenous NE acting at the autoreceptor.



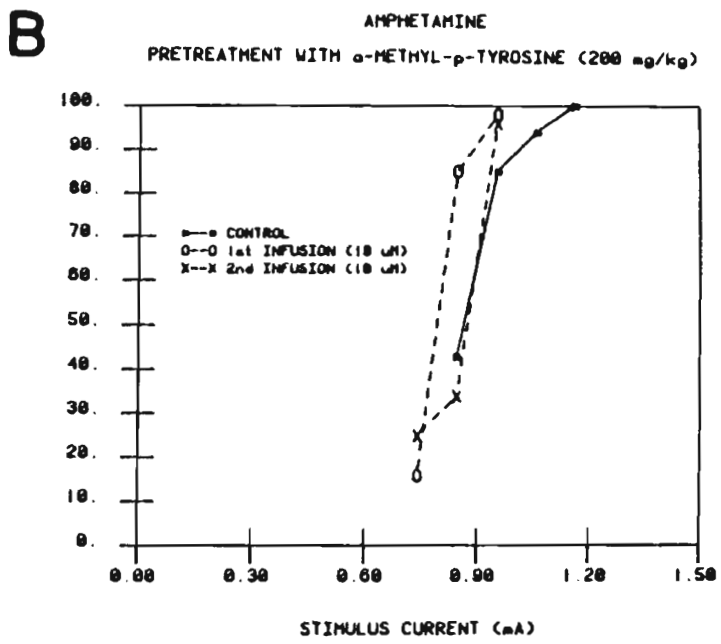


Fig. 5. Effects of local infusion of amphetamine upon the terminal excitability of locus coeruleus neurons. Infusion of 10 μM amphetamine into the terminal field in the frontal cortex produces decreased terminal excitability in a dose-related fashion. B. In animals depleted of NE by pretreatment with alpha-methyl-p-tyrosine (200 mg/kg, i.p.), the terminal excitability remains unchanged by 10 μM amphetamine infusion. (5A reprinted from Nakamura et al., 1982a, with permission.)

In marked contrast to local cortical infusion, intravenous injection of amphetamine in low doses (0.25-0.5 mg/kg) produces increased noradrenergic terminal excitability, illustrated in Figure 6 (Nakamura et al., 1982a; Ryan et al., 1985). This biphasic effect depending on the route of administration is due to differential effects on locus coeruleus neuron firing rates. The impulse activity of locus coeruleus neurons is known to be inhibited by the systemic injection of low-dose amphetamine, which acts indirectly via NE activation of alpha$_2$-somatodendritic autoreceptors (Engberg and Svensson, 1979; Graham and Aghajanian, 1971; Nakamura et al., 1982a; Ryan et al., 1985). Furthermore, there is some evidence that amphetamine's effect upon NE release is dependent on impulse traffic. Indeed, the amount of amphetamine-induced NE release from the terminal is much less in the absence of impulse traffic than in the presence of impulse flow along the axons (Raiteri et al., 1974). Therefore, when low-dose amphetamine is injected intravenously, the net amount of NE released from locus coeruleus terminals actually decreases

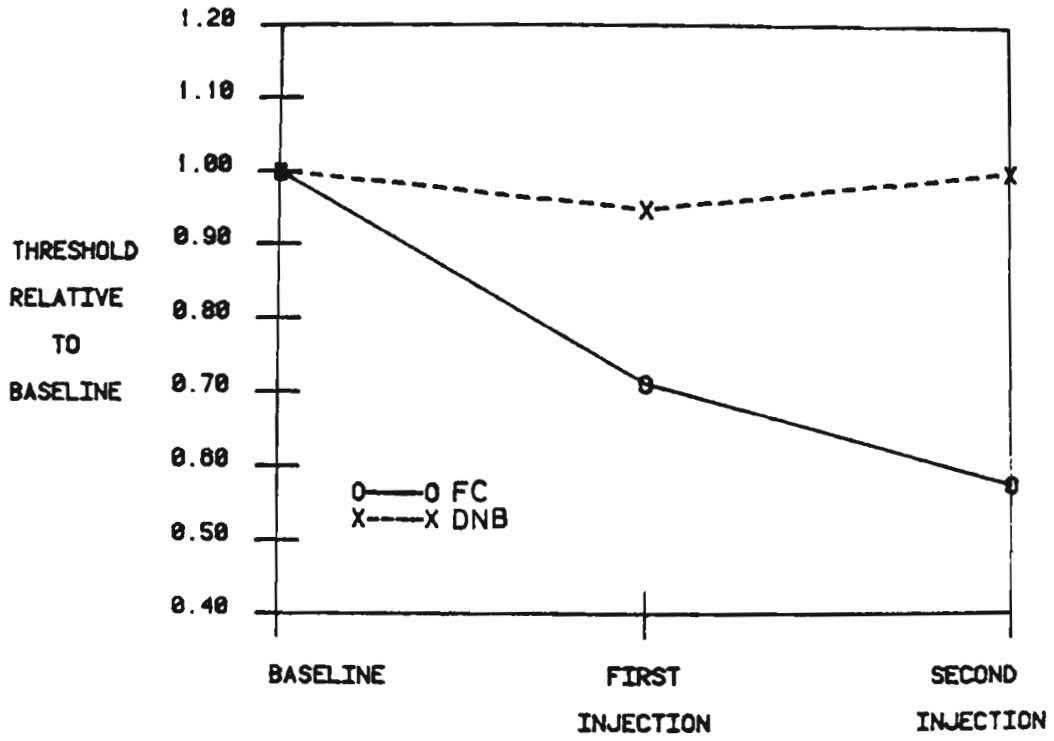


Fig. 6 Effects of low dose intravenous injection of amphetamine upon the excitability of noradrenergic terminals in the frontal cortex (FC) compared to that of the more proximal axon in the dorsal noradrenergic bundle (DNB). Two successive injections (0.5 mg/kg) produce a dose-related increase in the excitability of the terminal but not the pre-terminal axon. (From Nakamura et al., 1982a, with permission.)

as a result of the decreased impulse flow. Thus, terminal autoreceptors are less stimulated, and the terminal excitability increases. This interpretation is supported by low-dose amphetamine induced disinhibition of hippocampal cells by noradrenergic afferents, an effect that is abolished by a lesion of the locus coeruleus (Huang and Maas, 1981).

A high dose of amphetamine, administered intravenously, induced an effect opposite to that produced by a low-dose (Ryan et al., 1985). Despite a nearly complete suppression of impulse activity and a marked increase in terminal excitability induced by a prior low-dose of amphetamine, the terminal excitability of locus coeruleus neurons was markedly decreased by a subsequent intravenous injection of high-dose amphetamine. Both the low- and high-dose effects can be blocked by the alpha$_2$-antagonist, yohimbine. Therefore, at low doses, the ability of amphetamine to induce increased extraneuronal levels of NE at locus coeruleus terminals is dependent upon ongoing impulse activity. However, at higher doses the release of NE is largely independent of impulse flow, and terminal excitability is decreased due to increased noradrenaline release despite a complete inhibition of firing at the cell body (Nakamura et al., 1982a; Ryan et al., 1985a).

3.3. Effects of Impulse Traffic

The amount of neurotransmitter released per impulse from nerve endings is dependent upon the frequency of impulses reaching the terminals. If more NE is released within the terminal fields by increased impulse traffic, stimulation of the terminal autoreceptors by the endogenous ligand would increase. Similarly, periods of reduced impulse flow would lead to decreased autoreceptor stimulation as the extracellular concentration of transmitter in the region of the synapse falls. Therefore, the excitability of noradrenergic terminals might be expected to vary with changes in the spontaneous firing rate. This has been observed in locus coeruleus neurons in which the spontaneous firing rate is unstable: decreased terminal excitability is associated with periods of increased firing rate. Conversely, increased terminal excitability accompanies periods of decreased firing rate, as shown in Figure 7 (Nakamura et al., 1981).

Decreased terminal excitability, similar to that caused by the alpha$_2$-agonists and spontaneous increases in firing rate, can also be produced by artificially increasing the rate of impulses reaching the terminals (Nakamura et al., 1981). To increase the frequency of the impulse flow along noradrenergic axons, the dorsal noradrenergic bundle arising in the locus coeruleus was

electrically stimulated at threshold current at 10 Hz. After such stimulation, the cortical terminal excitability was decreased, whereas 10 Hz stimulation of the dorsal noradrenergic bundle by a subthreshold current only slightly lower than the threshold current did not cause any change in terminal excitability, as illustrated in Figure 8. In this series of experiments, it was also noted that increased impulse traffic produced a high frequency stimulation that had no effect on the excitability of the pre-terminal noradrenergic axons themselves in the dorsal noradrenergic bundle. Thus, the stimulation-induced changes in excitability are limited to the terminal fields of locus coeruleus neurons and do not represent non-specific alterations in the excitability of noradrenergic axons. These findings indicate that autoreceptor-mediated modulation of noradrenergic terminal excitability can occur in vivo, without application of exogenous drugs.

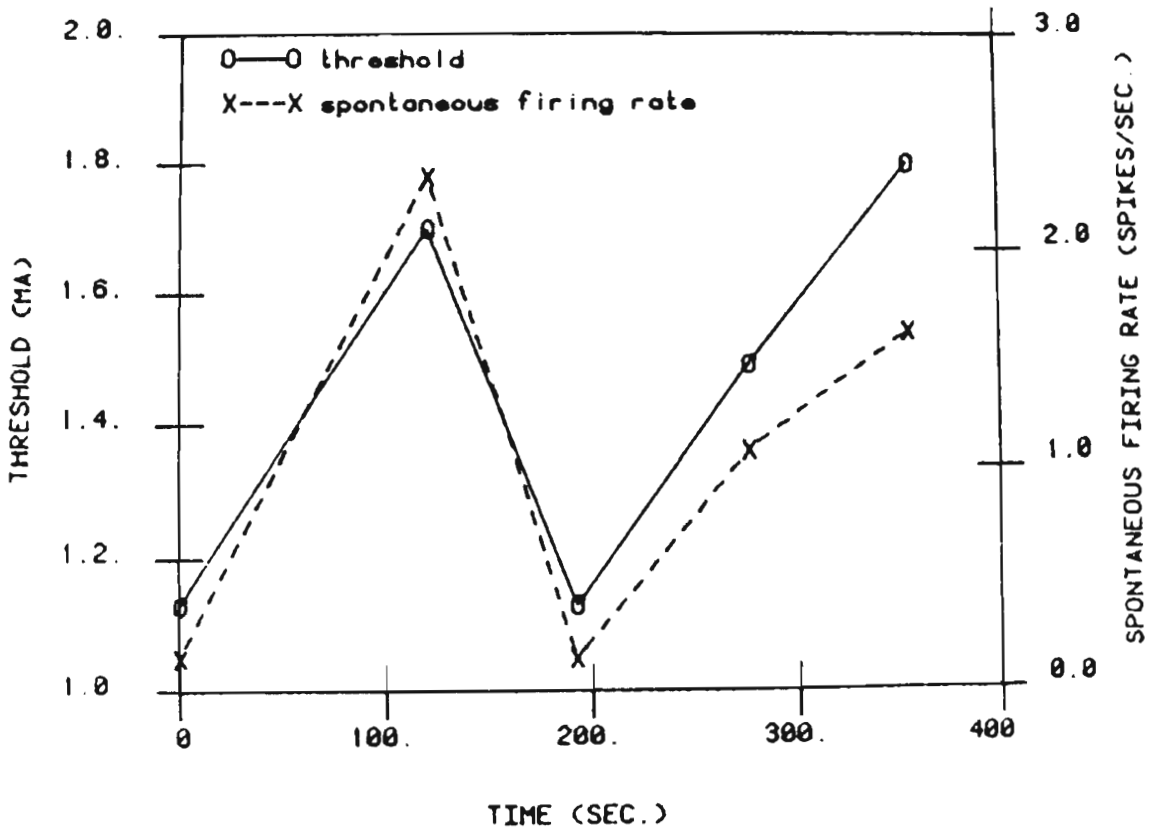


Fig. 7. Relationship between spontaneous firing rate and threshold currents necessary to evoke 100% antidromic invasion. Note that the threshold (left ordinate) varies with changes in spontaneous firing rate (right ordinate). (From Nakumara et al., 1981, with permission.)

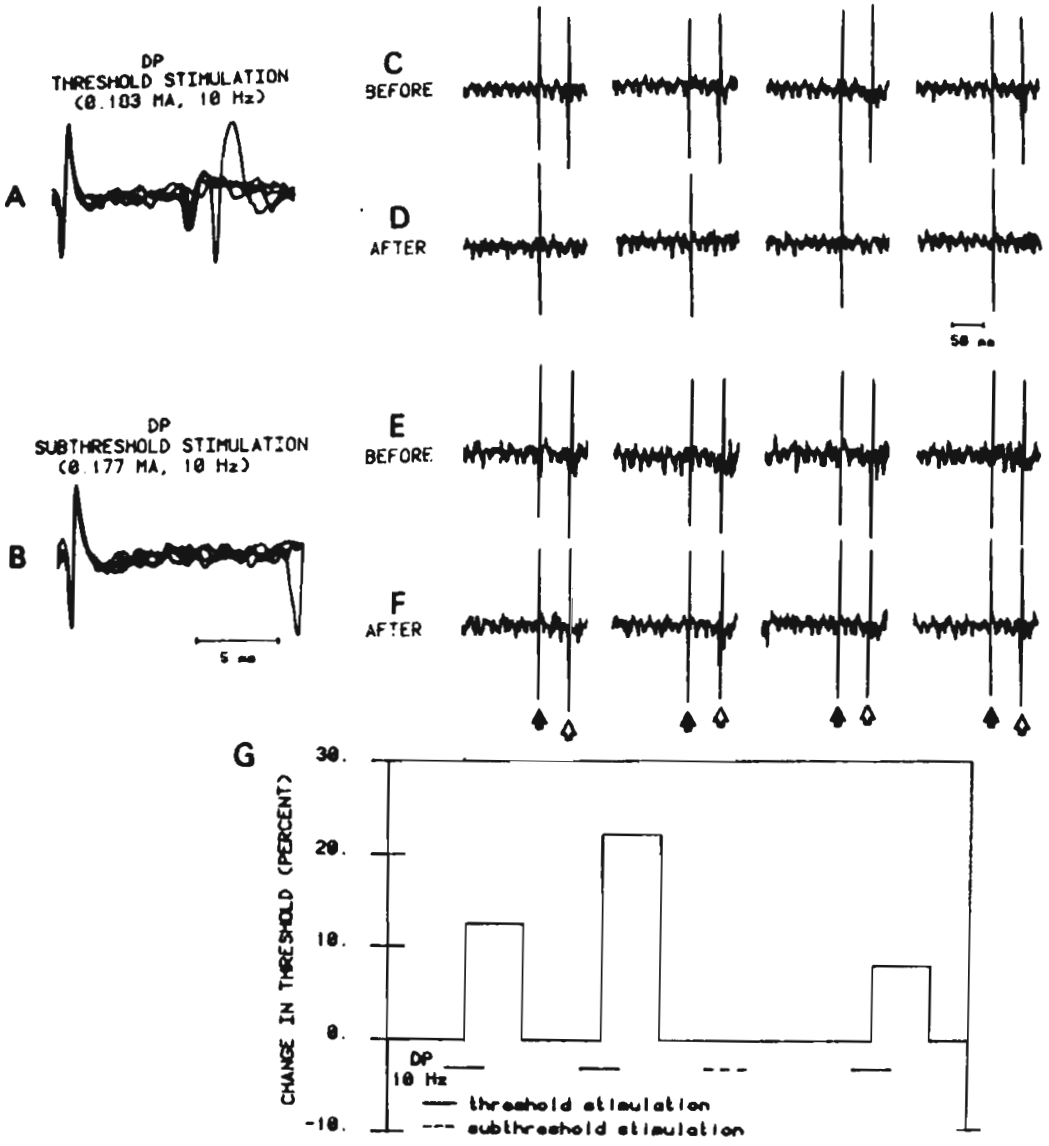


Fig. 8. High frequency stimulation of the dorsal noradrenergic bundle (DNB, 10 Hz, 10 s) produces a decrease in cortical noradrenergic terminal excitability. A. 10 Hz stimulation of the DNB effectively stimulates the axon as shown by the faithful appearance of the antidromic IS spike. C. Prior to stimulation of the DNB, cortical stimulus is at threshold as evidenced by an antidromic response to each stimulus. D. AFter 10 Hz stimulation of the DNB for 10 s, the control threshold current is unable to elicit any antidromic spikes, indicating decreased terminal excitability. B. If the DNB is stimulated at a slightly lower amplitude incapable of evoking antidromic action potentials, terminal excitability is unaffected (E, F). G. Summary of DNB effects for a typical locus coeruleus neuron. (From Nakamura et al., 1981, with permission.)

4. STIMULATION OF PRESYNAPTIC OPIATE RECEPTORS

4.1. Effects of Morphine and Opiate Peptides

4.1.1 Opiate Agonists

Biochemical studies of neurotransmitter release have demonstrated that like alpha$_2$-agonists, activation of presynaptic opiate receptors by morphine or opioid peptides causes a decrease in stimulus-evoked NE release from central noradrenergic nerve endings in vitro (Montel et al., 1974, 1975; Taube et al., 1977; Arbilla and Langer, 1978; Starke, 1980; Diez-Guerra et al., 1987; Werling et al., 1987). When infused directly into the frontal cortex, morphine (1–40 μM) or the stable synthetic opiate peptide analogue D-Ala, Met-enkephalinamide (ENK; 10 μM) results in a dose-related decrease in the terminal excitability of locus coeruleus neurons, without altering the neuronal firing rate, as illustrated in Figure 9 (Nakamura et al., 1982b). The decreased terminal excitability caused by opioid peptides, like that produced by alpha$_2$-agonists or amphetamine administration, is accompanied by an increase in antidromic latency and in latency variability.

4.1.2. Naloxone

The decreased noradrenergic terminal excitability induced by opiate agonists can be blocked by the administration of the opiate antagonist naloxone (10 μM), as shown in Figure 10 (Nakamura et al., 1982b). It was consistently observed that the effects of ENK were more easily antagonized by naloxone than were the effects of morphine. Intravenous injection and local infusion of naloxone reversed the decreased terminal excitability produced by a prior infusion of ENK (FIgure 10A), whereas naloxone exerted only a small antagonizing effect upon a prior infusion of morphine. However, if naloxone was infused into the terminal field prior to morphine infusion, the morphine-induced decreased terminal excitability was clearly attenuated (Figure 10B). The differential efficacy of naloxone on ENK- or morphine-induced decreases in noradrenergic terminal excitability could have resulted from a conformational change in the terminal opiate receptors induced by the prior morphine binding, or might indicate the presence of multiple opiate receptors on the noradrenergic nerve terminals. The latter is unlikely, however, since a recent biochemical study has indicated that only mu-type, but not delta- or kappa-type, opioid receptors mediate opiate-induced inhibition of NE release in rat cerebral cortex slices (Werling et al., 1987).

Administration of naloxone by itself resulted in increased terminal

excitability in some locus coeruleus neurons, though in many cells terminal excitability was not affected by the drug. Thus, at least some cortical noradrenergic terminals were under a tonic opioid inhibition in vivo.

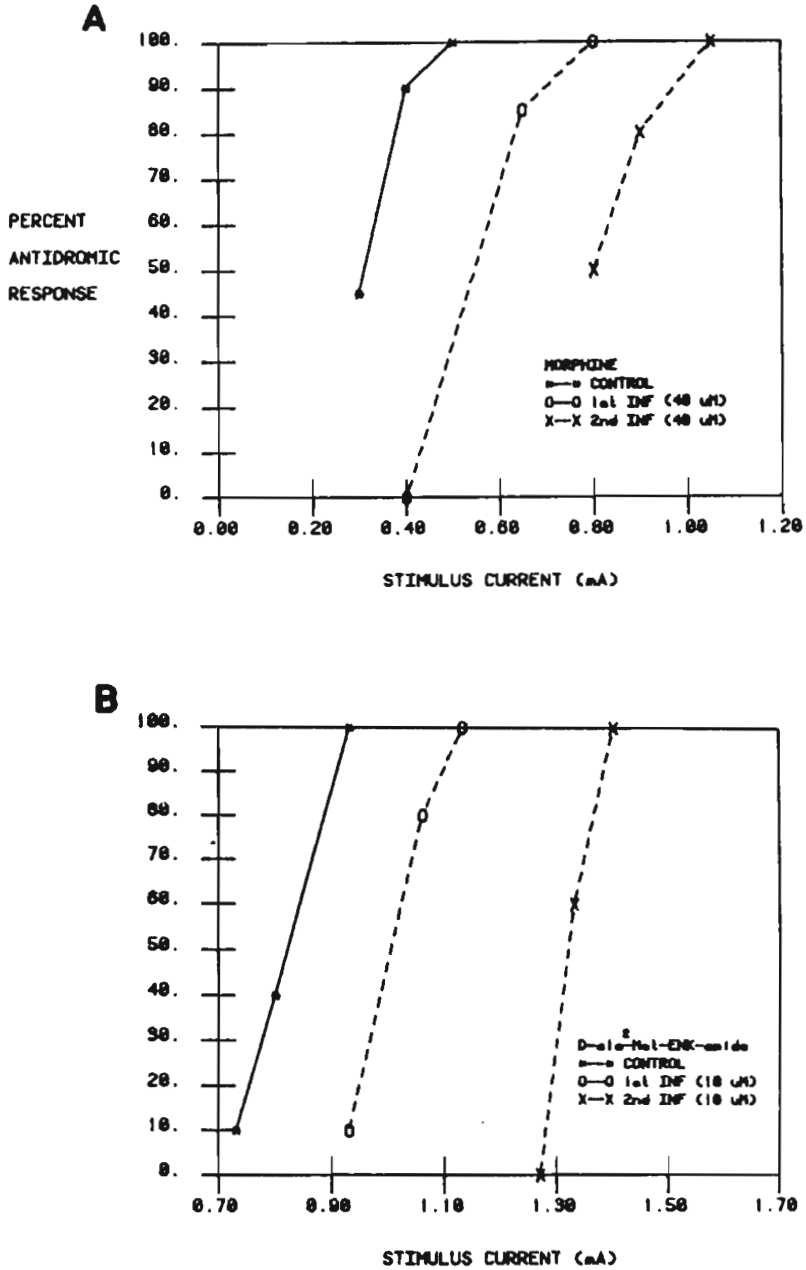


Fig. 9 Effects of local infusion of opioid agonists upon the terminal excitability of locus coeruleus neurons. A. Effects of morphine: Infusion of 40 μM morphine produces decreased terminal excitability. B. Effects of 10 μM D-Ala, Met-enkephalinamide (ENK), a stable synthetic opiate peptide analogue: Similar to the effects of morphine, ENK causes decreased terminal excitability in a dose-related manner. (9A redrawn from Nakamura et al., 1982b, with permission.)

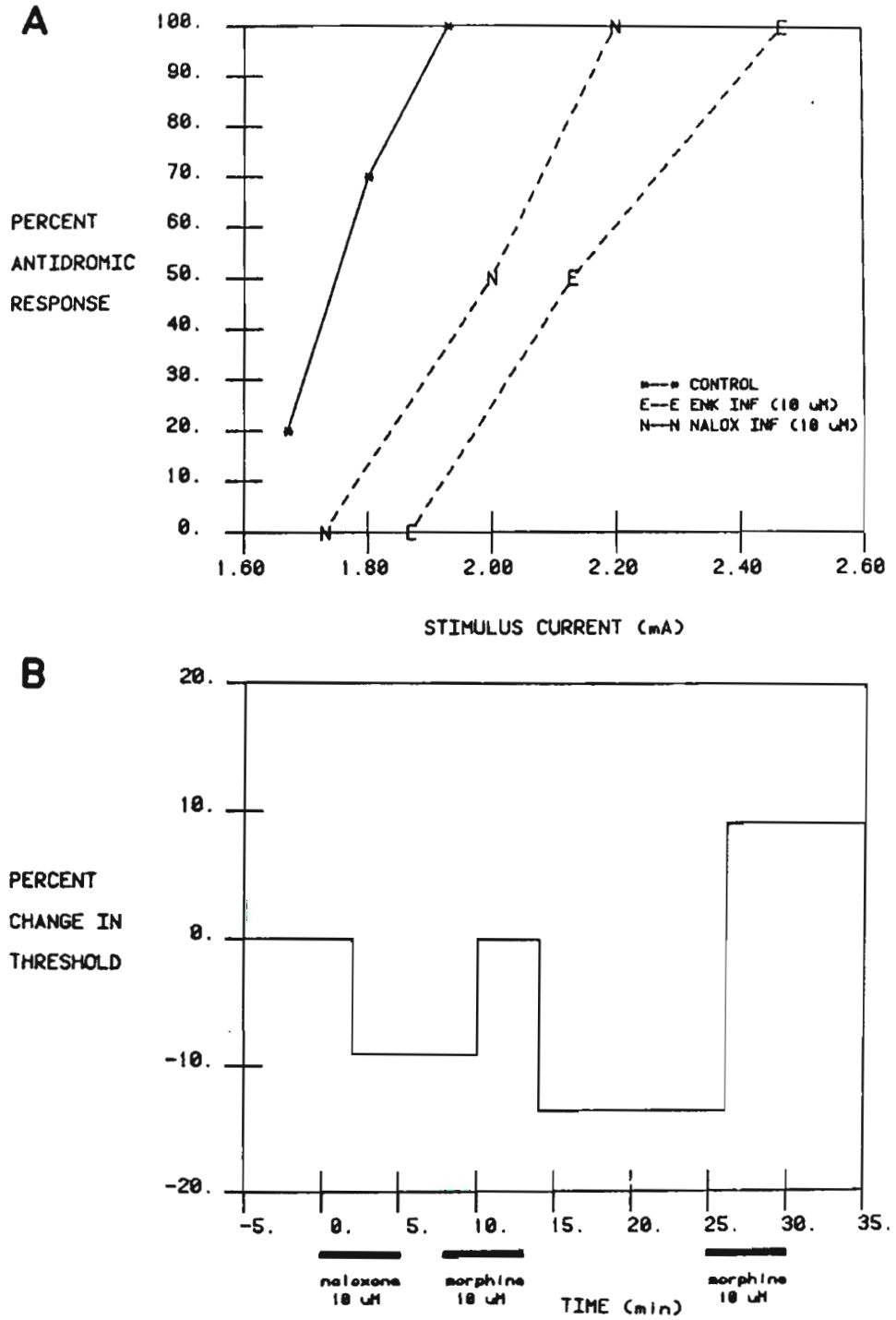


Fig. 10. Antagonistic effects of the opioid antagonist naloxone upon opioid agonists. A. Effects of naloxone upon ENK: Local infusion of naloxone partially reverses the decreased terminal excitability produced by prior infusion of ENK. B. Effects of naloxone upon morphine: Local infusion of naloxone by itself produces a slight increase in terminal excitability, as revealed by a small decrease in threshold for 100% antidromic invasion. Morphine infusion, immediately after naloxone infusion, shows only a small decrease (9%) in terminal excitability and the decreased terminal excitability quickly reverses following the cessation of morphine infusion. When morphine infusion is started long after naloxone infusion, a decrease in terminal excitability becomes much more noticeable (23%), and the change continues long after the cessation of morphine infusion. (10A redrawn from Nakamura et al., 1982b, with permission.)

The neurophysiological consequences of opiate agonist and antagonist administration into the terminal field is virtually identical to the alterations in terminal excitability produced by alpha$_2$-adrenergic agonists and antagonists. In addition, the administration of morphine and opiate peptides has been shown to inhibit the firing of locus coeruleus neurons, just as alpha$_2$-adrenergic agonists do (Bird and Kuhar, 1977; Guyenet and Aghajanian, 1979; Korf et al., 1974; Strahlendorf et al., 1980; Young et al., 1977). Furthermore, North and Williams (1985) have shown that the same potassium conductance is activated by both opiate and alpha-adrenergic agonists in the soma-dendritic region of locus coeruleus neurons hyperpolarizing the soma-dendritic membrane. Although no attempt has yet been made to examine the interaction between autoreceptors and opiate receptors located on noradrenergic axon terminals, it is likely that a common mechanism, as demonstrated for the soma-dendritic receptors, may underlie neurophysiological events resulting from stimulation of autoreceptors and opiate receptors on noradrenergic nerve terminals. This is discussed in greater detail in the next chapter, with respect to the actual mechanism(s) of transmitter release modulation.

5. PHYSIOLOGICAL SIGNIFICANCE OF PRESYNAPTIC OPIATE RECEPTORS ON NORADRENERGIC NERVE ENDINGS

Terminal autoreceptors may play a role in regulating the amount of NE released from the noradrenergic terminals in relation to impulse traffic (Nakamura et al., 1981). If more impulses reach the terminal fields per unit time, the stimulation of terminal autoreceptors will be increased as the extracellular concentration of NE increases, resulting in a decrease in further impulse-dependent NE release from the terminals. Conversely, less impulse activity at the terminals leads to an increase in subsequent impulse-dependent release by reducing activation of terminal autoreceptors. Therefore, autoreceptors located on the noradrenergic terminals may operate to maintain the amount of NE within the terminal field at a constant level by modulating the release of NE. The physiological significance of NE autoreceptors is fairly well established. However, there is some doubt as to the functional importance of opioid receptors localized on the terminals of NE neurons; particularly in light of the failure of morphological studies to provide evidence of axo-axonic synapses at cortical NE terminals (Olschowa et al., 1980). Consequently, some investigators have considered it unlikely that opiate receptor stimulation at NE terminals occurs in a manner analogous to

that of classical presynaptic inhibition (i.e. through the action of synaptic contact between neighboring axons (Nicoll and Alger, 1980). Moreover, no effective stimulus has been directly identified for the release of an endogenous opioid purportedly stimulating opioid receptors situated on NE nerve terminals. On the other hand, the failure to identify such a stimulus at present by no means precludes its existance. Furthermore, both intracellular and immunohistochemical studies in the locus coeruleus (LC) have revealed respectively 1) the presence of common α_2 and opioid affector mechanisms in the LC, and 2) the coexistance of endogenous opioid peptides and NE in this region (Aghajanian and Wang, 1987; Charnay et al., 1982; Leger et al., 1983). Moreover, the coexistence and the co-release of endogenous opioids and NE have also been demonstrated in both sympathetic terminals and in the adrenal medulla (Schuttzberg et al., 1978; Viveros and Wilson, 1983; De Potter et al., 1987). This evidence suggests the possibility that opioid receptors localized on NE cell bodies (e.g. the locus coeruleus) and terminals are activated by the release of endogenous opioids co-existing with NE in the neuron. The released opioid may therefore serve to regulate the impulse traffic of the NE neurons themselves. This proposal is strengthened by recent evidence that nanomolar amounts of endogenous opioid peptides can inhibit central NE release by stimulating opioid receptors localized on noradrenergic nerve terminals (Mulder et al., 1987). Further evidence for the physiological stimulation of terminal opioid receptors on NE neurons by an endogenous opioid peptide is provided by the finding that naloxone by itself often increases the terminal excitability of LC neurons in vivo. It is thus quite possible that, when released possibly together with NE, an opioid peptide coexisting with NE in noradrenergic neurons stimulates opioid receptors situated on NE cell bodies and terminals, resulting in an inhibition of NE release. This together with α_2 receptor stimulation may constitute a means of regulating (i.e. terminating) the release of NE once it has occurred. Further experiments, measuring terminal excitability in vivo, as has been described here, may be useful to explore the physiological conditions under which stimulation of these terminal opiate receptors takes place.

6. CONCLUSIONS

In this chapter we have described an in vivo method for examining the neurophysiological effects of the stimulation and blockade of presynaptic receptors located at the terminals of central noradrenergic neurons. Application of this method of terminal excitability testing to the cortical terminals of

central noradrenergic neurons of the locus coeruleus has revealed that autoreceptor and opiate receptor-induced decreases in the evoked release of NE in vitro are associated with decreases in terminal excitability in vivo. This decreased terminal excitability probably arises from a hyperpolarization of the presynaptic terminal, as we discuss further in the following chapter. Conversely, autoreceptor and opiate receptor antagonists lead to increases in terminal excitability in vivo, and increases in stimulated release of NE in vitro. Changes in noradrenergic terminal excitability also arise from changes in the rate of impulses reaching the terminal fields, suggesting a physiological role for autoreceptors in the regulation of noradrenergic neurotransmission in vivo. In the next chapter we will review the results of similar experiments conducted on central dopaminergic and serotonergic neurons, and discuss the physiological mechanisms underlying both the presynaptic receptor-mediated changes in monoamine terminal excitability and neurotransmitter release.

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