0022-3565/83/2243-0501$02.00/0
THE JOURNAL OF PHARMACOLOGY AND EXPERIMENTAL THERAPEUTICS

Vol. 224, No. 3
Printed in U.S.A.

Evidence for a Central Sympathoexcitatory Action of *Alpha*-2 Adrenergic Antagonists

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Accepted for publication November 29, 1982

ABSTRACT

The effect of *alpha*-2 adrenergic receptor antagonists on sympathetic nerve discharge (SND) recorded from the external carotid and splanchnic nerves were studied in baroreceptor-denervated cats. Low i.v. doses of piperoxane and rauwolscine dramatically increased SND and produced a concomitant rise in mean arterial pressure and heart rate. High doses of piperoxane also resulted in an increase in SND. High doses of rauwolscine, however, markedly reduced mean arterial pressure, heart rate and SND. The pressor response to i.v. norepinephrine was greatly attenuated by high doses of rauwolscine and slightly reduced by piperoxane. Piperoxane failed to alter SND in catecholamine-depleted animals. *In vitro* binding experiments indicated that piperoxane and rauwolscine bound selectively to the *alpha*-2 adrenergic receptor and had little affinity for *alpha*-1 receptor sites. In addition, rauwolscine displaced [^{3}H]LSD binding. These data indicate that low doses of the *alpha*-2 receptor antagonist piperoxane and rauwolscine act centrally to increase SND. In addition, the nature of the interactions between central noradrenergic neurons and neurons involved in the genesis of sympathetic nerve activity is discussed.

A growing body of evidence suggests that central noradrenergic neurons are intimately involved in cardiovascular control (Reis *et al.*, 1977; Loewy and McKellar, 1980). However, the nature of the interaction between central noradrenergic neurons and neurons involved in the genesis of SND remains uncertain. For example, some reports suggest that the noradrenergic input to central sympathetic neurons is excitatory (Neumayr *et al.*, 1974; Andén *et al.*, 1978; Loewy *et al.*, 1979; McCall and Humphrey, 1981), while others imply that the input is inhibitory (DeGroat and Ryall, 1967; Coote and Macleod, 1974; Coote *et al.*, 1981). Still others suggest that noradrenergic neurons excite brain stem components of baroreceptor pathways which results in a secondary decrease in SND (Haeusler, 1974; McCall and Gebber, 1976; Kobinger, 1978).

The centrally acting antihypertensive agent clonidine has been postulated to produce its effects by acting either as an *alpha*-2 adrenergic autoreceptor agonist or by mimicking the effects of norepinephrine at postsynaptic *alpha*-2 receptor sites. In the former case, clonidine has been shown to act at *alpha*-2 autoreceptor sites to decrease activity in central noradrenergic pathways (Schmitt *et al.*, 1971; Starke and Montel, 1973; Cedarbaum and Aghajanian, 1976, 1977). This implies that central noradrenergic neurons act directly to facilitate transmission in central sympathetic pathways or indirectly to antagonize an inhibitory input to sympathetic neurons. Conversely, if clonidine acts on postsynaptic *alpha*-2 adrenergic receptors, then noradrenergic neurons must act directly to inhibit sympathetic neurons or indirectly to excite inhibitory interneurons (*e.g.* baroreceptor interneurons).

Recent data from our laboratory indicate that the specific *alpha*-1 adrenergic receptor antagonists prazosin and WB-4101 act centrally to reduce SND (McCall and Humphrey, 1981). These observations are not consistent with the concept that central noradrenergic neurons act solely to inhibit sympathetic neurons. Rather, these data suggest that noradrenergic neurons facilitate central sympathetic outflow and that clonidine acts by decreasing activity in noradrenergic pathways (*i.e.*, disfacilitation). If true, selective *alpha*-2 receptor antagonists (*e.g.* piperoxane and rauwolscine) should increase central sympathetic outflow. In this regard, *alpha*-2 antagonists increase central norepinephrine turnover as well as the firing rate of norepinephrine-containing neurons (Andén *et al.*, 1976; Cedarbaum and Aghajanian, 1976; Miach *et al.*, 1978). However, high doses of piperoxane fail to increase SND (Schmitt *et al.*, 1971, 1973; McCall and Humphrey, 1982). We, therefore, have examined the effects of a broad range of i.v. doses of piperoxane and rauwolscine on SND. The results are compatible with the hypothesis that noradrenergic neurons facilitate central SND.

Received for publication July 27, 1982.

ABBREVIATIONS: SND, sympathetic nervous discharge; MAP, mean arterial pressure; HR, heart rate; LSD, lysergic acid diethylamide; 5-HT, serotonin.

Materials and Methods

General. Twenty-five cats weighing 2.0 to 4.0 kg were anesthetized by an i.p. injection of sodium diallylbarbiturate (60 mg/kg), urethane (240 mg/kg) and monoethylurea (240 mg/kg). The cats were placed in a David Kopf Instruments stereotaxic apparatus and a glass trachea tube was inserted. The femoral artery was cannulated to measure MAP with a Statham transducer and a Grass 7D polygraph. A femoral vein catheter was inserted to permit i.v. drug and vehicle administration. HR was recorded continuously with a Grass 7P4 tachograph triggered by the electrocardiogram (lead I–III). Rectal temperature was maintained between 36 and 38°C with a heating pad and/or lamp.

Baroreceptor denervation. The cats were baroreceptor denervated in order to accurately assess the effects of piperoxane and rauwolscine on SND. The carotid sinus, aortic depressor and vagus nerves were exposed from a ventral aspect after reflection of the cephalad portion of the trachea and esophagus into the mouth. The carotid sinus nerve was identified at its junction with the glossopharyngeal nerve, while the aortic depressor nerve was located at its junction with the superior laryngeal nerve. The vagus nerve was isolated in the midcervical region. Baroreceptor denervation was accomplished by bilaterally sectioning these nerves. The denervation was judged complete if SND was not inhibited during the pressor response to i.v. norepinephrine (0.1 μg/kg).

Sympathetic nerve recording. The sympathetic postganglionic external carotid nerve was isolated distal to its junction with the superior cervical ganglion, ligated and sectioned near the carotid sinus. Nerve potentials were recorded monophasically under mineral oil with a bipolar platinum electrode after capacity coupled preamplification (low and high half-amplitude responses at 1 and 500 Hz, respectively). Nerve activity was displayed on a Grass polygraph and quantitated using a Grass P710B cumulative integrator. In several experiments, SND was similarly recorded from the left sympathetic preganglionic greater splanchnic nerve following a retroperitoneal approach. These cats were artifically respired after immobilization with gallamine triethiodide (4 mg/kg i.v.). Pneumothoracotomy was performed to minimize untoward respiratory movements.

Drug treatments. MAP, HR and SND were allowed to equilibrate for 1 hr after baroreceptor denervation. Pretreatment values for MAP, HR and SND were averaged over the last 10 min of the equilibration period. Six vehicle control cats were then treated with an i.v. injection of distilled water. MAP, HR and SND were monitored over the ensuing 2 hr with supplemental vehicle being given at 15, 30, 45, 60 and 75 min.

The *alpha*-2 adrenergic receptor antagonists piperoxane and rauwolscine (Carl Roth, Karlsruhe, FRG) were dissolved in 0.5 mg/ml of distilled water and tested over a 90-min period. The starting dose of each compound was 0.025 mg/kg and the cumulative dose of each antagonist was doubled at 15- min intervals. The effects of these agents on MAP, HR and SND were determined 10 to 15 min after each dose. The pressor response to 0.1 μg/kg of i.v. norepinephrine was also determined immediately before each dose of piperoxane, rauwolscine or vehicle.

Monoamine depletion was achieved by pretreating animals with reserpine and the methyl ester of α-methyl-*p*-tyrosine. Reserpine (1 mg/kg i.p.) was administered approximately 24 hr before the experiment, whereas α-methyl-*p*-tyrosine (200 mg/kg i.v.) was given 2 hr before administration of piperoxane.

Receptor binding. The effects of *alpha*-2 antagonists on [^{3}H]clonidine, [^{3}H]prazosin and [^{3}H]LSD binding were studied by homogenizing a cat brain in 50 mM THAM-HCl buffer (pH 7.4) using a Polytron at setting no. 7 for 30 sec. The homogenate was centrifuged at 40,000 × *g* for 10 min and the supernatant discarded. The pellet was resuspended with the Polytron and the centrifugation repeated. The pellet was resuspended in THAM-HCl buffer and diluted to 100 times the original weight.

The incubation mixture contained 2 ml of homogenate, 0.1 ml of ^{3}H-ligand and 0.1 ml of vehicle, piperoxane or rauwolscine. Incubation was at 30°C for 30 min. Samples were filtered through Whatman GF/B filters. Incubation tubes were rinsed with 5 ml of buffer and the filters were washed three times with 5 ml of cold THAM-HCl buffer. Filters were transferred to liquid scintillation vials containing 15 ml of ACS (Amersham Corp., Arlington Heights, IL) liquid scintillation fluid and counted.

Specific binding was defined as the total bound ^{3}H-ligand less the amount not displaced by 1 μM clonidine for the [^{3}H]clonidine binding, 1 μM phentolamine for the [^{3}H]prazosin binding and 500 nM bromo-LSD for the [^{3}H]LSD binding. Scatchard analysis was performed using 12 different concentrations of the ^{3}H-ligand. K_i values for the *alpha*-2 antagonists were calculated using the method fo Cheng and Pursoff (1973).

Statistical analysis. Differences in MAP, HR and SND between the vehicle and drug-treated groups were analyzed for statistical significance by an unpaired *t* test. Both the absolute values and changes from pretreatment were evaluated. A probability level of $P \leq .05$ was presumed to reflect statistical significance. Values are espressed as mean ± S.E.

Results

Representative examples of the effects of the *alpha*-2 receptor antagonists piperoxane and rauwolscine on MAP, HR and SND are illustrated in figures 1 and 2. Administration of low doses of piperoxane (0.025–0.05 mg/kg i.v.) resulted in an immediate increase in MAP (20 mmHg) which largely returned to pretreatment values within 10 min. Like MAP, HR was not significantly altered 10 to 15 min after piperoxane administration. In contrast, low doses of piperoxane resulted in a significant sustained increase in splanchnic preganglionic (fig. 1) and external carotid postganglionic (table 1) SND. High doses of piperoxane (1.6 mg/kg i.v.) were associated with a slight but significant decrease in MAP and a small increase in HR. Splanchnic SND remained significantly elevated when compared with pretreatment values (fig. 1). Rauwolscine (0.025 mg/kg i.v.) significantly increased MAP, but had little effect of HR. Progressively higher doses of rauwolscine (0.05–1.6 mg/kg i.v.) were associated with dose-related decreases in MAP. A significant bradycardia was also observed after the 1.6 mg/kg dose of rauwolscine (table 1). Like piperoxane, low doses of rauwolscine dramatically increased SND. However, a high dose of rauwolscine (1.6 mg/kg i.v.) markedly reduced SND (fig. 2). In contrast, the vehicle control cats did not experience any change in MAP, HR or SND. The effects of the vehicle, piperoxane and rauwolscine on MAP, HR and SND are summarized in table 1.

Figure 3 illustrates the dose-response for the effects of vehicle, piperoxane and rauwolscine on MAP, HR, external carotid SND and the pressor response to i.v. norepinephrine (0.1 μg/kg i.v.). Low doses of piperoxane and rauwolscine were devoid of any effect on the norepinephrine challenge. High doses of piperoxane produced a slight attenuation in the response to norepinephrine (−23%) which coincided with a small drop in MAP. In contrast, high doses of rauwolscine markedly diminished the norepinephrine response (−71%). This effect was associated with substantial reductions in both MAP (−53%) and SND (−62%) and a slight bradycardia (−16%).

In order to establish that the increase in SND observed after piperoxane and low doses of rauwolscine resulted from their ability to antagonize *alpha*-2 adrenergic receptors, the effects of piperoxane in cathecholamine-depleted cats were studied. Animals received reserpine (1 mg/kg i.p.) 24 hr before drug testing and α-methyl-*p*-tyrosine (200 mg/kg i.v.) 2 hr before the start of the experiments. Figure 4 illustrates that piperoxane failed to significantly alter MAP, HR or SND in reserpine-pretreated cats ($n = 3$).

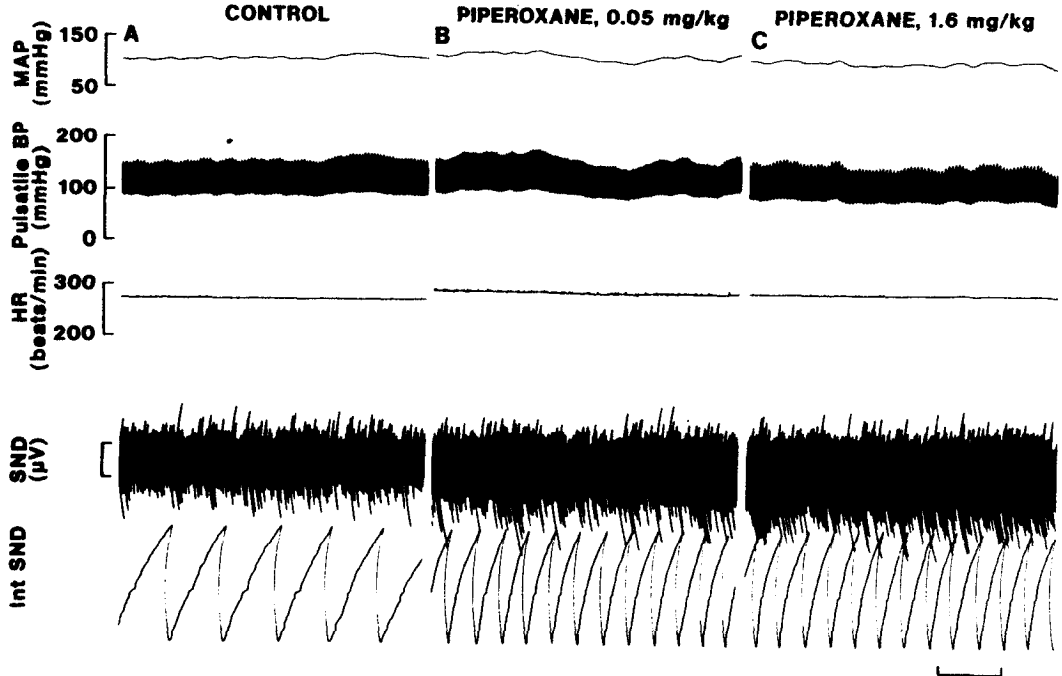

Fig. 1. Effect of piperoxane (0.05 and 1.6 mg/kg i.v.) in the baroreceptor-denervated cat. Tracings from top to bottom are MAP, pulsatile blood pressure (BP), HR, splanchnic SND and integrated (Int) SND. Piperoxane markedly increased SND. Vertical calibration is 60 μV. Horizontal calibration is 1 min.

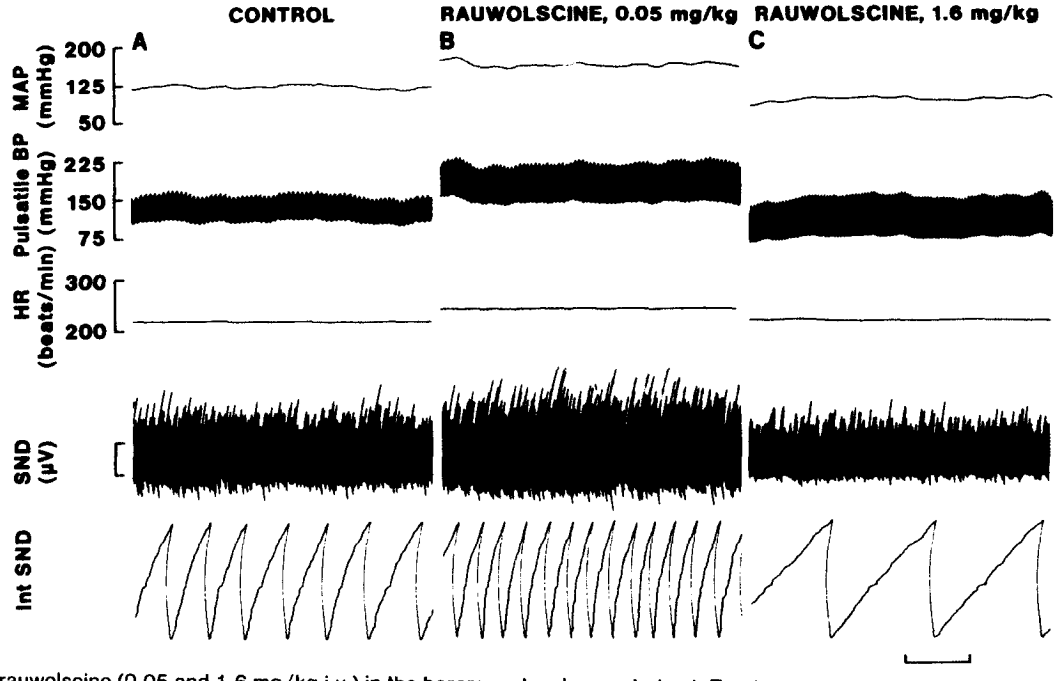

Fig. 2. Effect of rauwolscine (0.05 and 1.6 mg/kg i.v.) in the baroreceptor-denervated cat. Tracings from top to bottom are MAP, pulsatile blood pressure (BP), HR, external carotid SND and integrated (Int) SND. Rauwolscine significantly increased SND at low doses but reduced nerve activity at high doses. Vertical calibration is 40 μV. Horizontal calibration is 1 min.

The higher doses of rauwolscine closely resembled the established actions of the selective *alpha*-1 antagonist prazosin, which likewise causes hypotension, diminished SND and norepinephrine blockade (McCall and Humphrey, 1981). Inasmuch as these data suggest that rauwolscine might exhibit *alpha*-1 receptor antagonism at higher doses, the effects of rauwolscine and piperoxane on *in vitro alpha*-1 and *alpha*-2 adrenergic receptor binding were examined. The results are summarized in table 2. The data indicate that both these agents selectively bind to *alpha*-2 receptor sites in the cat brain. Rauwolscine was particularly effective in displacing [^{3}H]clonidine binding. Neither rauwolscine nor piperoxane had significant *alpha*-1 adrenoreceptor binding activity. However, rauwolscine did display modest affinity for the [^{3}H]LSD receptor.

TABLE 1

Effect of piperoxane and rauwolscine on MAP, HR and SND in baroreceptor-denervated cats

All values are expressed as mean ± S.E. PT, pretreatment.

Dose	Vehicle Control (n = 6)			Piperoxane (n = 6)			Rauwolscine (n = 7)		
	MAP	HR	% SND	MAP	HR	% SND	MAP	HR	% SND
mg/kg									
PT	123 ± 6	231 ± 10		110 ± 10	241 ± 12		116 ± 10	226 ± 6	
0.025	125 ± 7	231 ± 10	103 ± 3	114 ± 10	250 ± 9	134 ± 8*	144 ± 8*	236 ± 8	148 ± 15*
0.05	123 ± 7	237 ± 8	102 ± 2	110 ± 12	252 ± 10	142 ± 12*	120 ± 8	237 ± 8	152 ± 14*
0.10	121 ± 6	232 ± 10	104 ± 3	104 ± 8	257 ± 11	147 ± 11*	115 ± 6	238 ± 9	170 ± 15*
0.20	122 ± 5	236 ± 10	110 ± 4	107 ± 9	262 ± 10	158 ± 13*	103 ± 10	244 ± 9	172 ± 19*
0.40	120 ± 6	240 ± 12	111 ± 6	108 ± 8	257 ± 12	166 ± 14*	96 ± 12	245 ± 8	144 ± 20
0.80	120 ± 6	241 ± 13	116 ± 6	111 ± 7	259 ± 13	162 ± 14*	64 ± 10*	231 ± 9	84 ± 14
1.6	121 ± 7	241 ± 13	114 ± 7	90 ± 5*	259 ± 13	156 ± 16*	55 ± 8*	190 ± 9*	38 ± 16*

* Indicates a significant difference ($P \leq .05$) from PT values.

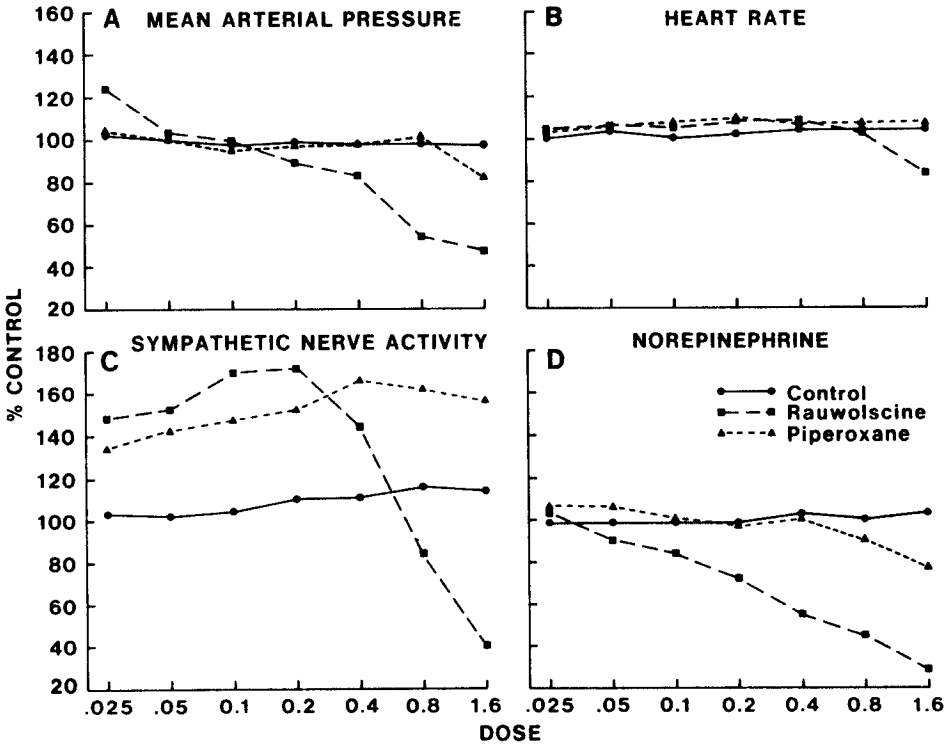

Fig. 3. Log dose-response curve of the effects of piperoxane, rauwolscine and vehicle on MAP, HR, SND and the pressor response to i.v. norepinephrine (0.1 μg/kg).

Discussion

The present investigation was designed to determine if the *alpha*-2 adrenergic receptor antagonists piperoxane and rauwolscine act in the central nervous system to increase SND. We found that low doses of piperoxane and rauwolscine markedly increased sympathetic activity recorded from the external carotid and greater splanchnic nerves of the baroreceptor-denervated cat. Whereas Laubie and Schmitt (1980) previously reported an increase in SND with piperoxane, these authors attributed this to an excitatory action at sympathetic ganglia. However, because piperoxane (fig. 1) and rauwolscine increased SND recorded from the preganglionic splanchnic nerve and postganglionic external carotid nerve to a similar degree, it appears that an excitatory effect of *alpha*-2 antagonists at sympathetic ganglia is negligible. That the splanchnic nerve is preganglionic in character is indicated by the lack of effect of ganglionic blockers on splanchnic SND. Taken together, these observations indicate that low doses of piperoxane and rauwolscine act centrally to increase sympathetic nerve activity. This sympathoexcitatory effect likely results from their *alpha*-2 receptor antagonist activity, the only known pharmacologic property common to these structurally dissimiliar agents. In addition, a third *alpha*-2 adrenoreceptor antagonist, yohimbine, potentiates the electrodermal and nictitating membrane reflexes in the cat (Koss and Bernthal, 1979). Thus, at least three *alpha*-2 adrenergic antagonists have been shown to increase sympathetic activity. Both *in vivo* and *in vitro* studies in the rat indicate that piperoxane and rauwolscine are specific *alpha*-2 antagonists in low doses (Anden *et al.*, 1976; Drew, 1976; Cedarbaum and Aghajanian, 1977; Lambert *et al.*, 1978; Miach *et al.*, 1978; Hedler *et al.*, 1981; Shepperson *et al.*, 1981). Our binding studies confirm that these agents are selective for *alpha*-2 receptor sites in the cat (table 2). In addition, we found that piperoxane failed to alter SND in cathecholamine-depleted animals (fig. 4). Taken together, these observations strongly suggest that piperoxane and rauwolscine increased SND as a consequence of their ability to antagonize *alpha*-2 adrenergic receptors.

The increased SND produced by piperoxane was sustained

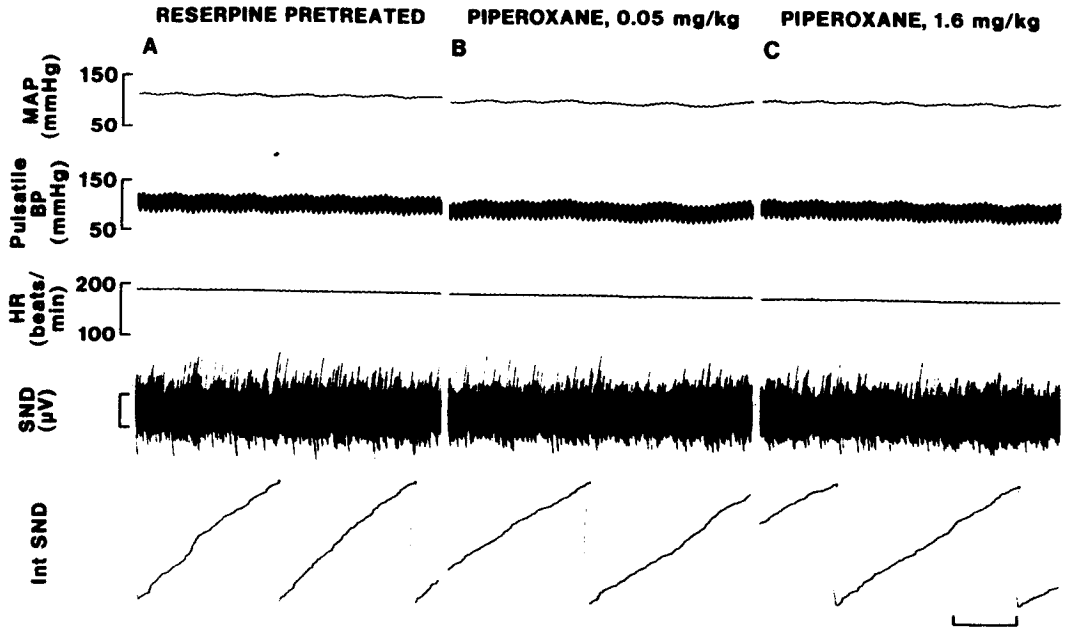

Fig. 4. Effect of piperoxane (0.05 and 1.6 mg/kg i.v.) in the reserpine pretreated, baroreceptor-denervated cat. Tracings from top to bottom are MAP, pulsatile blood pressure (BP), HR, external carotid SND and integrated (Int) SND. Piperoxane failed to alter SND in catecholamine-depleted animals. Vertical calibration is 60 μV. Horizontal calibration is 1 min.

TABLE 2

Inhibition of specific [^{3}H]clonidine and [^{3}H]prazosin binding to crude cat brain membrane preparation by adrenergic compounds

Drug	IC_{50}			K_i		
	[^{3}H]Clonidine	[^{3}H]Prazosin	[^{3}H]LSD	[^{3}H]Clonidine	[^{3}H]Prazosin	[^{3}H]LSD
	M	*M*	*M*	*nM*	*nM*	*nM*
Piperoxane	1.05×10^{-7}	1.11×10^{-6}	3.65×10^{-7}	49.5	186	243
Rauwolscine	6.1×10^{-9}	1.34×10^{-6}	1.75×10^{-8}	2.9	224	11.6
Clonidine				3.5		
Prazosin					0.36	

at high doses and was associated with a small decrease in MAP and the pressor response to i.v. norepinephrine and a minimal change in HR. The decrease in MAP in the presence of an increase in SND suggests that piperoxane must have additional effects on blood pressure regulating mechanisms. In contrast, high doses of rauwolscine significantly reduced MAP, HR, SND and the norepinephrine pressor response (fig. 3). This suggested that high doses of rauwolscine might possess a significant *alpha*-1 adrenergic receptor blocking action. Subsequent binding studies, however, indicated that neither rauwolscine nor piperoxane had appreciable activity at *alpha*-1 adrenergic receptor sites (table 2). Rauwolscine did significantly bind to LSD receptors (table 2) and has been shown to inhibit serotonin (5-HT)-induced contractions of the rat fundus (Lambert *et al.*, 1978). These observations suggest that rauwolscine may have significant 5-HT receptor antagonist activity. Because we have previously demonstrated that 5-HT antagonists act centrally to reduce MAP, HR and SND (McCall and Humphrey, 1982), this property of rauwolscine may explain the decreases in MAP, HR and SND observed at high doses of this drug. On the other hand, *in vitro* assessments of selectivity for *alpha* adrenoreceptor blocking agents are not always reflected under *in vivo* conditions (Massingham *et al.*, 1981). Thus, it is possible that *in vivo* rauwolscine may have significant *alpha*-1 receptor antagonist activity in the cat. This may in part explain the pronounced depression of the norepinephrine pressor response seen after high doses of rauwolscine. In this regard, other investigators have found that the *alpha*-2/*alpha*-1 receptor antagonist ratio of rauwolscine *in vivo* is only 10/1 to 30/1 (Madjar *et al.*, 1980; Shepperson *et al.*, 1981). In addition, the inhibition of the pressor response after piperoxane and rauwolscine may be related to the fact that the pressor response to exogenously administered norepinephrine is mediated in part by *alpha*-2 adrenergic receptors (Yamaguchi and Kopin, 1980). Clearly, further studies are required to determine the nature of the inhibition of SND and the norepinephrine pressor response after high doses of rauwolscine.

The present study may shed additional light on the role of central noradrenergic neurons in the control of blood pressure. There are at least three classes of adrenergic compounds which markedly alter sympathetic outflow from the central nervous system. First, the *alpha*-2 receptor agonist clonidine produces a profound depression of SND (Schmitt *et al.*, 1971, 1973; McCall and Gebber, 1976; Kobinger, 1978). Second, *alpha*-1 receptor antagonists such as prazosin and WB-4101 also reduce SND (McCall and Humphrey, 1981). Finally, as shown in the present investigation, the *alpha*-2 antagonists piperoxane and rauwolscine increase SND. Any comprehensive theory on the nature of the interaction between noradrenergic neurons and central sympathetic pathways must take these three observations into account. The fact that *alpha*-1 adrenergic antagonists act centrally to reduce SND implies that 1) central noradrenergic neurons normally facilitate sympathetic outflow and 2) clonidine acts at *alpha*-2 autoreceptors to reduce noradrenergic

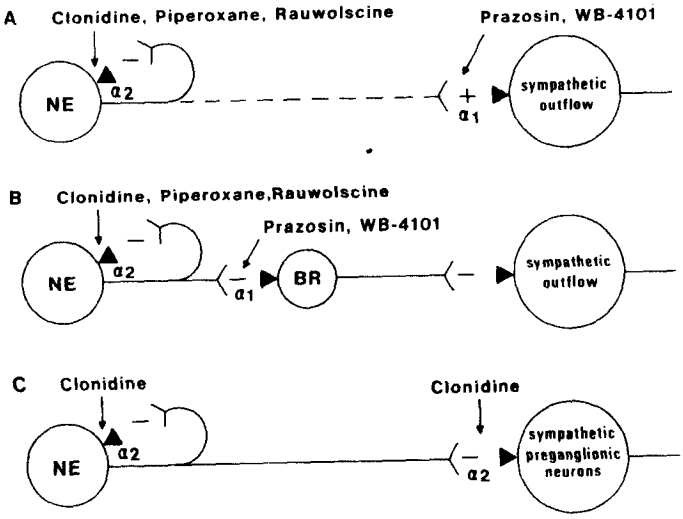

Fig. 5. Models of the role of noradrenergic (NE) neurons in the control of sympathetic outflow from the central nervous system and possible sites of action of adrenergic agents. A, NE neurons excite central sympathetic neurons directly or indirectly (dashed line). Clonidine acts at *alpha*-2 (α_2) receptors located on NE neurons to inhibit the firing of these neurons. Prazosin blocks the excitatory action of NE at *alpha*-1 (α_1) receptors. B, NE neurons inhibit interneurons [*e.g.*, baroreceptor (BR) neurons] which normally inhibit central sympathetic neurons. Clonidine reduces SND by acting at NE neurons to disinhibit BR interneurons. Prazosin antagonizes the inhibitory action of NE on the inhibitory interneuron. C, NE neurons inhibit sympathetic preganglionic neurons *via* an *alpha*-2 receptor. The net effect of i.v. clonidine results from inhibition of spinal preganglionic neurons and excitation *via* an inhibition of NE firing.

firing, thus inhibiting SND by disfacilitation. This is illustrated by the models in figure 5. Noradrenergic neurons may act either directly to excite central sympathetic neurons (fig. 5A) or indirectly to antagonize inhibitory neurons (*e.g.* baroreceptor interneurons) which project to sympathetic neurons (fig. 5B). In either case, the effect of norepinephrine would be mediated *via* an *alpha*-1 receptor.

On the other hand, it has been demonstrated that clonidine can reduce SND in cathecholamine-depleted animals (Haeusler, 1974; Kobinger and Pichler, 1976). This suggests that endogenous norepinephrine is not required for the central cardiovascular action of clonidine and that the action of clonidine is mediated by postsynaptic *alpha*-2 adrenergic receptors. In support of this contention, Guyenet and Cabot (1981) found that sympathetic preganglionic neurons are inhibited by microiontophoretically applied clonidine. The inhibition was mediated by an *alpha*-2 adrenoreceptor (fig. 5C). Together, these data suggest that noradrenergic inputs to central sympathetic neurons are inhibitory. The net effect of i.v. clonidine would result from direct inhibition of central sympathetic neurons and excitation *via* an inhibition of noradrenergic neuronal firing (*i.e.*, disinhibition; fig. 5C).

The contradictory conclusions drawn from the effects of *alpha*-1 receptor antagonists and spinal clonidine suggest that noradrenergic neurons might produce opposite effects on sympathetic neurons in different areas of the central nervous system. In support of this contention, *alpha*-1 receptor antagonists typically reduce SND by a maximum of 40%, whereas clonidine inhibits SND by at least 80% (McCall and Humphrey, 1981). In addition, although clonidine reduces SND in cathecholamine-depleted animals, the dose required for inhibition is at least three times greater than in control animals (Haeusler, 1974). This suggests that both pre- and postsynaptic *alpha*-2 receptors are important in mediating the central sympatholytic action of clonidine. Thus, clonidine may act directly to inhibit sympathetic neurons (fig. 5C) and indirectly at *alpha*-2 autoreceptors to inhibit noradrenergic firing. This would result in a disfacilitation of an excitatory action on sympathetic neurons (fig. 5A) or a disinhibition of sympathoinhibitory interneurons (fig. 5B). In contrast, prazosin would act only at *alpha*-1 receptors to antagonize the action of norepinephrine (fig. 5, A and B). The *alpha*-2 adrenergic antagonists likely increase SND by acting postsynaptically to block the direct inhibitory effects of norepinephrine of sympathetic neurons and by increasing the firing of noradrenergic neurons (*i.e.*, a presynaptic receptor action). Obviously, additional studies are required to establish the sites of action of *alpha*-1 and *alpha*-2 antagonists as well as clonidine. In addition, the action of these agents on central epinephrine-containing pathways needs to be determined. In this regard, epinephrine neurons project to relevant cardiovascular areas of the brain (Hokfelt *et al.*, 1973) and likely mediate their effects *via alpha*-2 adrenergic receptors (Franz *et al.*, 1982).

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