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Cocaine Modulation of Central Monoaminergic Neurotransmission

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PITTS, D. K. AND J. MARWAH. *Cocaine modulation of central monoaminergic neurotransmission.* PHARMACOL BIOCHEM BEHAV 26(2) 453–461, 1987.—Extracellular microelectrode studies were conducted to test the effects of cocaine HCl on the activity of spontaneously firing single serotonergic dorsal raphe (DRN), noradrenergic locus coeruleus (LC) and dopaminergic ventral tegmental (VTA) and zona compacta (ZC) neurons, and cerebellar Purkinje neurons (PC) in urethane anesthetized rats *in vivo*. Cocaine (0.0625–4 mg/kg) predominantly inhibited all of the central monoaminergic neurons and predominantly activated cerebellar Purkinje neurons. Cocaine (1 mg/kg, IV) *failed* to potentiate the inhibitory effects of LC stimulation on PC neurons. The temporal effects of intravenous cocaine on arterial pressure (i.e., pressor response) were *not* directly correlated with the effects on neurons. Cocaine did *not* decrease the amplitude or slope of neuron action potentials, and the effects of cocaine on firing rate were *not* shared by similar doses of procaine. Reserpine pretreatment (10 mg/kg, IP) attenuated the effects of cocaine (1 mg/kg, IV) on DRN, LC, and PC neurons. Specific adrenoceptor antagonists antagonized the inhibitory effects of cocaine on LC (piperoxane, yohimbine) and VTA (haloperidol) neurons. These results suggest that the central effects of cocaine on presynaptic monoaminergic neurons may in part be mediated by augmented monoamine neurotransmission at autoreceptors and that the effects of cocaine on postsynaptic target cells (PC) may be more complex, requiring the analysis of both pre- and postsynaptic elements.

Cocaine In vivo Monoaminergic neurotransmission

IN the United States, cocaine abuse is a continuing and a growing problem [19]. Cocaine is both a local anesthetic and a psychoactive drug [26]. It has complex and numerous actions on the central nervous system. Many of the psychotropic effects of cocaine are considered to arise as a consequence of its interactions with central monoaminergic systems rather than its local anesthetic properties [8, 23, 25, 33]. Supporting this contention are biochemical studies, indicating that cocaine inhibits the uptake of monoamine neurotransmitters [16, 17, 27, 29] and perhaps causes their release from monoaminergic neurons [27,28]. Such effects of cocaine would markedly prolong the time that monoamine transmitters spend in the synaptic cleft. As a consequence, the synaptic actions of these transmitters would be exaggerated. Although a large body of behavioral [5, 9, 13, 30–32] and biochemical [6, 12, 16, 28, 29] evidence indicates that the psychotropic effects of cocaine are elicited by augmented central monoaminergic neurotransmission, there currently exists only one report [25] of the effects of this drug on single identified spontaneously firing central monoaminergic neurons. This is an important consideration since such studies would shed light on the fundamental neurologic mechanisms/perturbations that contribute to the psychotropic actions of cocaine. In addition such studies may indicate a strategy for more effective pharmacotherapeutic intervention.

This manuscript represents an evaluation of the acute effects of cocaine and the structurally related local anesthetic, procaine, on the spontaneous activity of single noradrenergic neurons in the nucleus locus coeruleus (LC), single serotonergic neurons in the dorsal raphe nucleus (DRN), single dopaminergic neurons in the substantia nigra-zona compacta (ZC) and the ventral tegmental area (VTA) and single spontaneously firing cerebellar Purkinje neurons (PC). This detailed study provides evidence which suggests that cocaine exerts profound effects on central monoaminergic neurotransmission and provides an insight into its mechanistic basis of action at the single neuron level.

METHOD

General

Male Sprague-Dawley rats (Charles River), weighing 170—290 g, used in these studies were maintained on a 12/12 hour light-dark cycle with food and water provided ad lib. The animals were anesthetized with urethane (1.25 g/kg, IP), intubated after a tracheotomy and allowed to breathe spontaneously. Body temperature was monitored by a rectal probe and maintained at 37±1°C by a heating pad. A catheter was inserted into the lateral tail vein for the administration of drugs or the vehicle, physiological saline.

Electrophysiology

The animals were mounted in a Kopf stereotaxic instru-

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ment for electrophysiological recording and a 3 mm bur hole was drilled in the skull overlying the area of interest. Stereotaxic coordinates used for extracellular single monoaminergic neuron recording were: locus coeruleus (LC—A6) 1.2 mm posterior to lambda and 1.1 mm lateral to midline; ventral tegmental area (VTA—A10) 3.0 anterior to lambda and 0.5 mm lateral from midline; zona compacta (ZC—A9) 3.0 mm anterior to lambda and 2.0 mm lateral to midline; and dorsal raphe (DRN—B7) 0.5 mm anterior to lambda at midline. Cerebellar Purkinje neuron recordings were made in the vermis, lobules VI and VII within an area with maximal depth approximately 1 mm from the brain surface.

Neurons encountered during electrophysiological recording were tentatively identified as monoaminergic when they met the following criteria: locus coeruleus—(1) electrical activity of noradrenergic LC neurons usually encountered at a depth of approximately 5.0–6.0 mm below the skull surface often with a zone of electrical silence ventral to the cerebellum and above the LC corresponding to the fourth ventricle; (2) a characteristic positive-negative long duration (~2 msec) action potential, usually with a notch on the ascending limb; (3) the presence of the mesencephalic nucleus of cranial nerve V (trigeminal) just lateral to the LC recording area, whose cells are readily activated by moving the lower mandible of the rat; (4) a firing rate of 0.5–5.0 Hz; (5) a characteristic acceleration of firing rate followed by inhibition when pressure was applied to the contralateral hindpaw; (6) a consistent, rapid and reversible inhibition produced by small (~10 μg/kg) systemic doses of clonidine.

Serotonergic neurons in the dorsal raphe were recognized as: (1) positive-negative spikes with a characteristic regular rhythm and (2) slow rate (0.5–2.5 Hz) appearing in the midline just ventral to an area of electrical silence (cerebral aqueduct). Dorsal raphe neurons were usually encountered (3) 5–5.5 mm ventral to the dura. Such neurons had a (4) wide duration of action potential (1–2 msec), were usually encountered as "doublets" or "triplets" and were (5) exquisitively sensitive to the inhibitory effects of the intravenous administration of LSD (~10 μg/kg, IV).

Dopaminergic neurons of the ventral tegmental area and substantia nigra, zona compacta area were located (1) 6.5–7.5 mm ventral to the cortical surface at the coordinates given above. These cells usually displayed a (2) long duration positive-negative action potential (~2 msec) frequently associated with a prominent after potential and a (3) slow (2–8/sec) bursting, complex spike pattern in which there is a progressive decrease in spike amplitude within each action potential cluster. These neurons were (4) significantly inhibited by apomorphine (~10 μg/kg, IV) and accelerated by haloperidol (~50 μg/kg, IV).

Cerebellar Purkinje neurons were identified by their (1) characteristic discharge of complex and simple negative-positive spikes.

Only one cell was studied in each animal for all neuron types investigated.

The extracellular electrical activity of single neurons was recorded with a glass (Radnoti-quikfil) micropipette having a tip diameter of approximately 1 μ. The micropipette was filled with 2 M NaCl saturated with fast green (in vitro impedance 2.0–4 megaohm measured at 135 Hz) and lowered into the brain with a hydraulic microdrive. Each spike was amplified, discriminated and fed into an integrator (reset every 10 sec) and finally displayed on a storage oscilloscope. The output from the discriminator-integrator was also monitored on a chart recorder and an audiomonitor. Further processing of the window discriminator output (1, 2 or 10 second bins) was accomplished by a computer, which also generated neuron rate histograms. Only extracellular potentials having a signal (>150 μV) to noise (<30 μV) ratio of at least 5 were considered acceptable. Additional details of the electrophysiological recording procedure can be obtained from previous reports [20–22, 25].

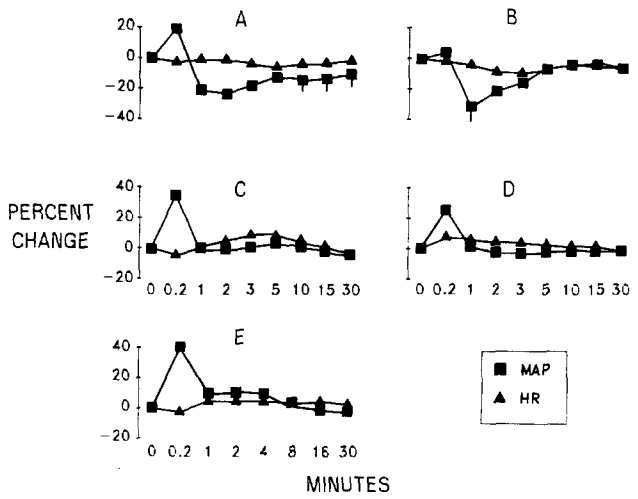

FIG. 1. Effect of cocaine or procaine on mean arterial blood pressure and heart rate under different anesthetic conditions. Ordinate scales represent percent change from baseline following intravenous administration of either 1.25 mg/kg cocaine (panels on left side; A, C, E) or 1.25 mg/kg procaine (panels on right side; B, D) and are the same from left to right. The abscissa scales represent time in minutes following cocaine administration and are the same for panels A–D. Panels A (n=5) and B (n=4) show the effects of cocaine and procaine in pentobarbital anesthetized rats. Panels C (n=4) and D (n=4) show the effects of cocaine and procaine in conscious-restrained rats. Panel E illustrates the effects of cocaine (1 mg/kg) in urethane anesthetized rats.

Stimulation of the locus coeruleus source of Purkinje neuron noradrenergic innervation was accomplished with a concentric bipolar electrode (Rhodes, 0.4 mm dia.) stereotaxically inserted into the ipsilateral cerebellar hemisphere. Ten-second trains of 0.2 msec pulses at 10 Hz were applied at two-minute intervals. Each experiment included 5 pre-drug or control stimulation trials followed by 5 post-drug stimulation trials.

Cardiovascular Recordings

Catheters were placed in the left femoral artery. These catheters consisted of 60 cm lengths of polyethelene tubing (PE 50), which were filled with heparinized saline (Sigma, 40 units/ml) and connected to a Gould Statham P23Db strain-gauge transducer [24]. The output of the transducer was conditioned by a Gould pressure processor and displayed on a Gould chart recorder and digital display as heart rate and mean, systolic, diastolic and pulse pressures.

Data Analysis

Data analysis was accomplished using the two-sample *t*-test, paired *t*-test or the Mann-Whitney test [34]. Data is expressed as mean±standard error unless otherwise indi-

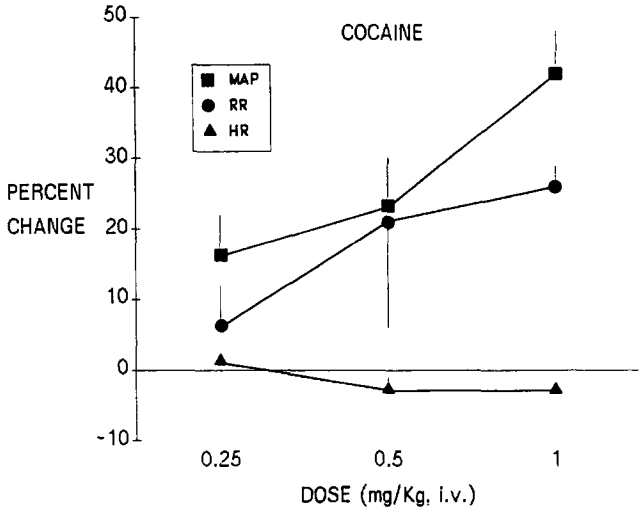

FIG. 2. Dose related effects of intravenous cocaine on MAP (squares), respiratory rate (circles) and heart rate (triangles) at the time of the maximum increase in MAP (~15 seconds) in urethane anesthetized rats. The ordinate represents the percent change from baseline for the three cardiovascular parameters. The abscissa represents the dose of cocaine administered on a log scale (0.25 mg/kg, n=6; 0.5 mg/kg, n=6; 1 mg/kg, n=6).

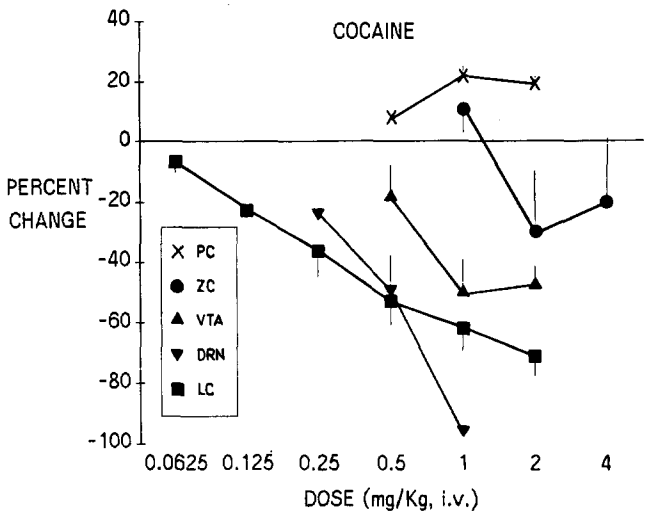

FIG. 3. Dose related effects of cocaine on five different types of CNS neurons: cerebellar Purkinje cells (PC), dopaminergic zona compacta cells (ZC), dopaminergic ventral tegmental area cells (VTA), serotonergic dorsal raphe cells (DRN), and noradrenergic locus coeruleus cells (LC) two minutes after intravenous administration. Ordinate represents percent change from a two-minute baseline period. Abscissa represents cocaine doses on a log scale (expressed as 0.0625–4 mg/kg). See Table 1 for further details.

TABLE 1

RESPONSES* OF CENTRAL NEURONS TO COCAINE

Neuron Type	Dose (mg/kg)	% Excited	% Inhibited	% Biphasic	% No Effect	n
LC	0.0625	—	50	—	50	4
	0.125	—	100	—	—	6
	0.25	—	100	—	—	8
	0.5	—	100	—	—	9
	1	—	100	—	—	12
	1 (R)†	30	40	—	30	10
	2	—	100	—	—	6
DRN	0.25	—	100	—	—	3
	0.5	—	100	—	—	7
	1	—	100	—	—	6
	1 (R)†	75	25	—	—	8
VTA	0.5	—	60	—	40	5
	1	—	100	—	—	6
	2	—	100	—	—	4
ZC	1	40	20	—	40	5
	2	25	75	—	—	4
	4	40	60	—	—	5
PC	0.5	80	—	—	20	5
	1	75	17	8	—	12
	1 (R)†	14	—	—	86	7
	2	73	9	—	18	11

*Responses categorized by determining percent change in activity from a two minute baseline period relative to two minutes after drug administration. Excitation: >5% increase; Inhibition: >5% decrease; Biphasic: inhibition/excitation; No effect: ±5% (inclusive).

†Reserpine pretreatment, 10 mg/kg 5 hours before electrophysiological recording.

cated. A p value of less than 0.05 was considered significant in all statistical tests.

Drugs

The following drugs were utilized: yohimbine HCl (Sigma); cocaine HCl (Sigma); procaine HCl (Sigma); mepivacaine HCl (Winthrop); clonidine HCl (Boehringer Ingelheim); piperoxane HCl (Rhone-Poulenc); naloxone HCl (Endo); LSD (NIDA); apomorphine HCl (Sigma); haloperidol (McNeil); reserpine (Sigma). Reserpine (10 mg/kg) was administered intraperitoneally 5 hours before the commencement of electrophysiological recordings [11].

RESULTS

Preliminary experiments were conducted to assess the impact of intravenous cocaine on the cardiovascular system of anesthetized rats. Figure 1 shows the effects of similar doses of cocaine and the structurally related local anesthetic procaine on cardiovascular parameters measured under different anesthetic conditions. Panel A depicts the effects of 1.25 mg/kg cocaine on mean arterial blood pressure and heart rate in pentobarbital anesthetized rats. The predominant cardiovascular effect observed was a biphasic pressor/depressor response with minimal effects on heart rate. The pressor response was very brief with a mean onset following intravenous cocaine administration of 15±1 seconds. The pressor effect was followed by a more prolonged depressor effect. The local anesthetic, procaine (1.25 mg/kg, IV), also produced a prolonged depressor response as was observed with cocaine in the pentobarbital anesthetized animals (panel B). In sharp contrast, however, neither cocaine nor procaine elicited a depressor response in conscious-restrained rats (panels C and D respectively). Both cocaine and procaine did elicit brief pressor responses (duration of 16±2 and 8±3 seconds respectively). The effects of cocaine (1 mg/kg, IV) on mean arterial pressure and heart rate in urethane anesthetized rats (1.25 g/kg, IP, panel E) were generally very similar to those observed in conscious-restrained rats (panel C).

The cardiovascular effects of cocaine in urethane anesthetized rats were dose dependent (Fig. 2). In this dose range (0.25–1 mg/kg, IV) cocaine also elicited significant tachypnea.

The predominant effects of systemic (IV) cocaine on the central monoaminergic neurons studied was a prolonged inhibition of firing rate. The mean percent change in the activity of PC, LC, DRN, VTA and ZC neurons for various doses of cocaine is depicted in Fig. 3. All LC neurons were inhibited at doses above the threshold (0.0625 mg/kg dose, Table 1). Additionally, all DRN neurons were inhibited by the doses of cocaine employed (0.25–1 mg/kg, Fig. 3 and Table 1). In contrast to LC and DRN neurons, the effects of cocaine on the dopaminergic neurons appeared to be less consistent and less pronounced. Doses greater than 0.5 mg/kg were required to inhibit all the VTA neurons studied; and doses of cocaine as high as 4 mg/kg failed to inhibit all the ZC neurons studied (see Fig. 3, Table 1 and Fig. 5—panels C, D and E).

In contrast to the predominant inhibitory effect of cocaine on monoaminergic neurons the predominant effect of cocaine on cerebellar Purkinje neurons was activation (Fig. 3—depicts excitatory effects only). This excitatory effect of cocaine was small in magnitude relative to the inhibitory effects of cocaine on monoaminergic LC, DRN and VTA neurons, but this effect occurred with the greatest frequency

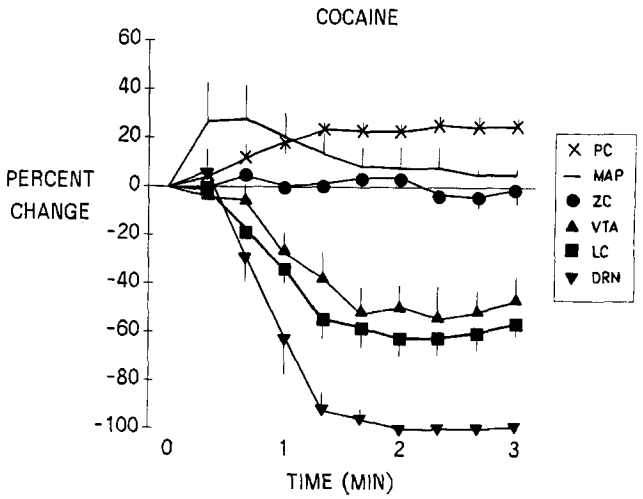

FIG. 4. Time course for the effects of intravenous cocaine (1 mg/kg) on MAP and central activity of single neurons (PC, n=7; ZC, n=6; VTA, n=5; LC, n=12; DRN, n=6). Ordinate represents percent change from baseline and abscissa represents time in minutes following drug administration.

at the doses of cocaine employed (0.5–2 mg/kg, Table 1).

None of the inhibitory effects of cocaine on the monoaminergic neurons or the activation effects on Purkinje cells appeared to be directly correlated with changes in cardiovascular parameters. Figure 4 shows that the effects of cocaine on central neurons persist after the brief pressor effect of cocaine is over. The maximal effect of cocaine on neuron firing was usually evident two minutes after drug administration, when the effects of cocaine had generally reached a plateau (Fig. 4).

Figure 5 illustrates typical effects of cocaine on each of the cell types studied. In panel A, although prior saline administration was without any significant effects on firing rate (not shown), subsequent administration of cocaine (1 mg/kg, IV) rapidly inhibited the spontaneous activity of a LC neuron. The specific opiate (mu) receptor antagonist naloxone (1 mg/kg, IV) did not reverse the inhibitory effects of cocaine; however, subsequent administration of the specific alpha-2-adrenoceptor antagonist, piperoxane (250 μg/kg, IV) rapidly and significantly reversed the effects of cocaine. Panel B shows the effects of saline and cocaine (1 mg/kg) on the spontaneous activity of a DRN neuron. This cell was completely inhibited in less than two minutes after drug administration (at 1 mg/kg 5 out of 6 DRN neurons were completely inhibited by cocaine). After some recovery from the effects of cocaine, this cell was silenced by the administration of LSD (20 μg/kg, IV, recovery began approximately two minutes later, data not shown). The inhibitory effects of cocaine on dopaminergic VTA neurons is illustrated in panels C and D. The neuron in panel C represents a neuron relatively sensitive to intravenous cocaine (1 mg/kg), while the neuron in panel D represents a relatively insensitive VTA neuron. Both neurons, however, were exquisitively sensitive to the inhibitory effects of the specific dopaminergic agonist, apomorphine (12.5 and 25 μg/kg, IV for panels C and D respectively), which were rapidly reversed by the subsequent administration of haloperidol (250 and 500 μg/kg, IV for panels C and D respectively). The dopaminergic ZC neuron depicted in panel E was excited by cocaine (1 mg/kg)

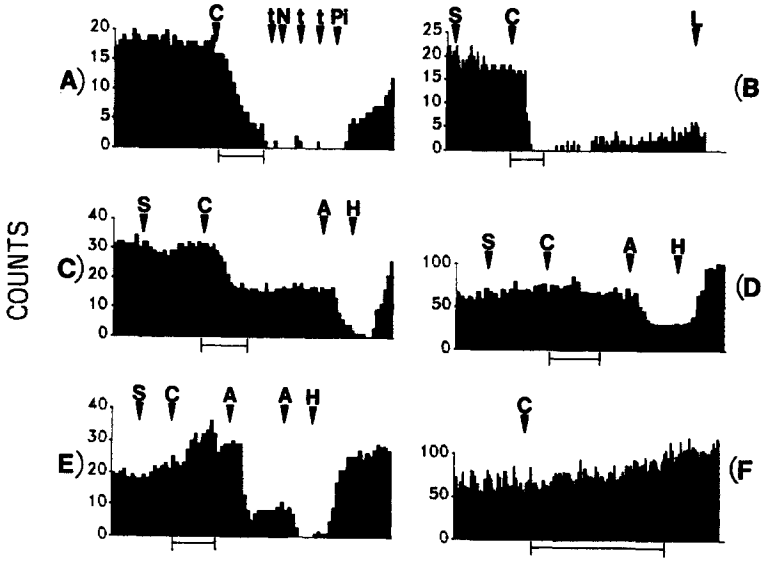

FIG. 5. Rate meter histograms depicting the effects of intravenous cocaine on six different single identified spontaneously firing neurons. The ordinates of panels A–E represent counts per 10 seconds and counts per 2 seconds in panel F. Each of the calibration bars represent a two minute period. Panel A=noradrenergic LC neuron; panel B=serotonergic DRN neuron; panel C=dopaminergic VTA neuron; panel D=dopaminergic VTA neuron; panel E=dopaminergic ZC neuron; panel F=cerebellar PC neuron. Intravenous drugs or treatments: C=cocaine, 1 mg/kg (panels A–F); S=physiological saline (panels B–E); t=toe pinch (nociceptive stimulus, panel A); Pi=piperoxane, 250 μg/kg (panel A); L=LSD, 20 μg/kg (panel B); A=apomorphine, 10 μg/kg ×2 (panel E), 12.5 μg/kg (panel C) and 25 μg/kg (panel D); H=haloperidol, 75 μg/kg (panel E), 250 μg/kg (panel C), and 500 μg/kg (panel D).

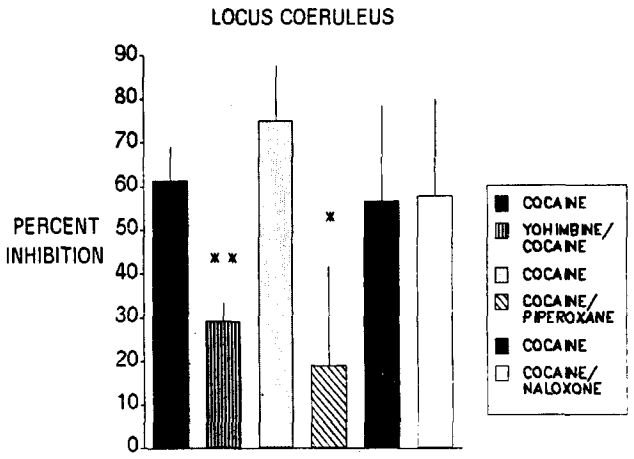

FIG. 6. Bar graph illustrating the effects of various antagonists on the inhibition of LC neurons elicited by the intravenous administration of 1 mg/kg cocaine. Data represents mean percent inhibition two minutes after cocaine administration. Cocaine, n=12; yohimbine pretreatment (5 mg/kg, IP) + cocaine n=6; cocaine; cocaine + piperoxane (250 μg/kg, IV), n=5; cocaine; cocaine + naloxone (1 mg/kg, IV), n=3. $^{**}p<0.01$, two-sample t-test (cocaine vs. yohimbine + cocaine). $^{*}p<0.05$, paired t-test (cocaine vs. cocaine + piperoxane, n=5).

administration. Subsequent administration of apomorphine (10 μg/kg, IV × 2) silenced this cell. Haloperidol (75 μg/kg, IV) reversed the inhibitory effects of apomorphine. A rate histogram for the activity of a cerebellar Purkinje neuron is shown in panel F. Prior saline administration was without significant effects on this cell (not shown). Cocaine (1 mg/kg, IV) elicited an activation of this neuron within approximately 1 minute.

The effects of various antagonists on the inhibition elicited by cocaine (1 mg/kg) for LC neurons is shown in Fig. 6. Pretreatment of animals with the specific alpha-2-adrenoceptor antagonist, yohimbine (5 mg/kg, IP), significantly attenuated the effects of subsequent cocaine administration on LC neurons ($p<0.01$). The specific alpha-2-adrenoceptor antagonist, piperoxane (250 μg/kg, IV), reversed the inhibitory effects of prior cocaine administration on 5 LC neurons ($p<0.05$). Naloxone (1 mg/kg, IV) did *not* affect the inhibition produced by prior cocaine administration (mean percent difference=1.4±4.1; $p>0.20$, n=3). In Fig. 7 haloperidol reversed (actually increased rate above baseline in all three animals, mean percent increase=242±78) the inhibitory effects of cocaine on dopaminergic VTA neurons ($p<0.05$).

As seen in Fig. 8, the ester linked local anesthetic, procaine, did *not* have any significant effects on the firing rate of noradrenergic LC neurons in cumulative doses up to 4 mg/kg, IV ($p>0.20$, when compared to mean percent inhibition of 1.8±2.6 in saline controls, n=17). The rate histogram from a single LC neuron is depicted in Fig. 9A. Cumulative doses of procaine up to 8 mg/kg did not significantly affect spontaneously firing rate of this neuron. These doses of procaine did, however, reduce mean arterial pressure

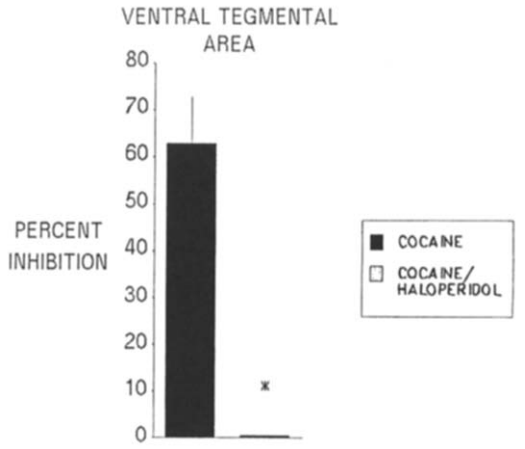

FIG. 7. Bar graph illustrating the effects of intravenous haloperidol (0.5 mg/kg, IV) on the inhibition of LC neurons elicited by the intravenous administration of 1 mg/kg cocaine, $p<0.05$, paired t-test (n=3).

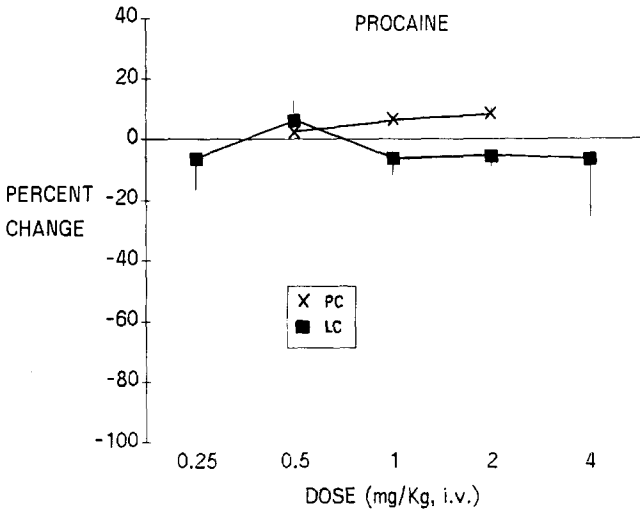

FIG. 8. Dose related effects of cumulative intravenous doses of procaine on LC (0.25–4 mg/kg, n=4) and PC neurons (0.5–2 mg/kg, n=4). See Fig. 3 for further details.

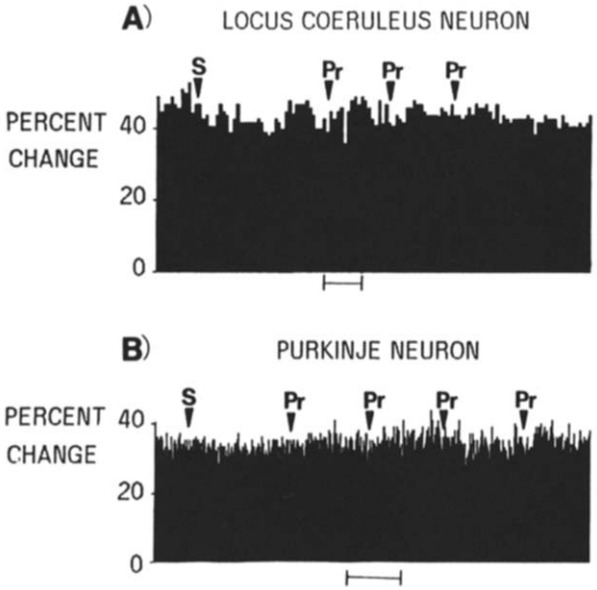

FIG. 9. Rate meter histograms depicting the effects of cumulative doses of procaine on a single LC neuron (panel A; S=saline, Pr=procaine: 2, 4 and 8 mg/kg, IV) and a single PC neuron (panel B; S=saline, Pr=procaine: 0.5, 1, 2, and 4 mg/kg, IV). The ordinates represent either counts per 10 seconds (LC) or counts per 1 second (PC). Each calibration bar represents a two minute period.

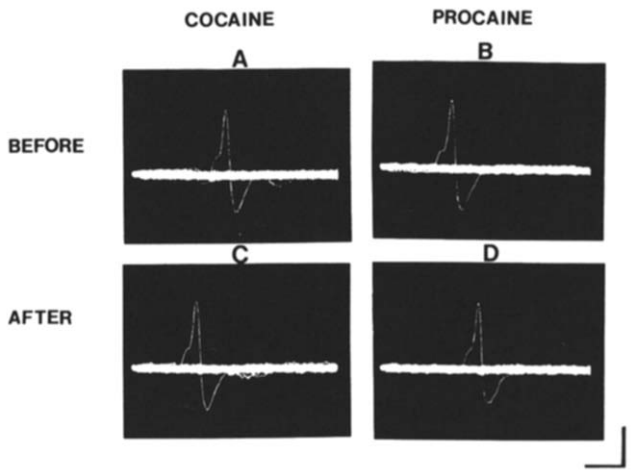

FIG. 10. Action potentials from two different single LC neurons before and after either cocaine (left column, 1 mg/kg, IV) or procaine (right column, 12 mg/kg, IV). Vertical calibration=200 μV; horizontal calibration=2 msec.

(~20%, data not shown). Cumulative doses of procaine did elicit a very small increase in activity of spontaneously firing PC neurons, however, cocaine produced a significantly greater increase in activity than procaine ($p<0.05$, Figs. 8 and 3). The rate histogram of a single, spontaneously firing PC neuron is shown in Fig. 9B.

Figure 10 illustrates the effects of cocaine (panels A and C) and procaine (panels B and D) on LC neuron action potential amplitude and waveform. Likewise Fig. 11 illustrates the effects of cocaine and procaine on Purkinje neuron action potentials. In neither case did cocaine or procaine affect action potential amplitude or slope.

Stimulation of the LC decreased the activity of postsynaptic spontaneously discharging PC neurons. The computer averaged response of 5 different control stimulus trials in 5 different animals is shown in Fig. 12A. The overall average inhibition of spontaneous PC activity was 52.1±6.1% prior to cocaine administration. Panel B shows a computer averaged response of LC induced inhibition of PC activity after the intravenous administration of 1 mg/kg cocaine in the same 5 animals as depicted in panel A. Cocaine did *not* potentiate the effects of LC stimulation on PC activity. The overall average inhibition of PC activity was significantly lower than that observed prior to drug administration (37.4±7.1%, $p<0.05$).

Figure 13 compares the time course for the effects of

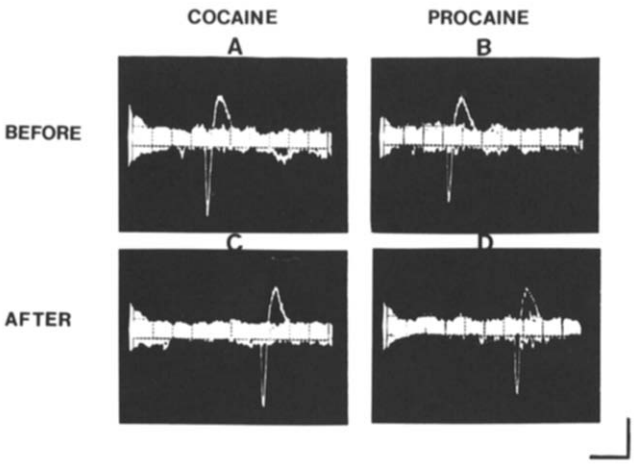

FIG. 11. Action potentials from two different single PC neurons before and after either cocaine (left column, 1 mg/kg, IV) or procaine (right column, 2 mg/kg, IV). Vertical calibration=100 μV; horizontal calibration=2 msec.

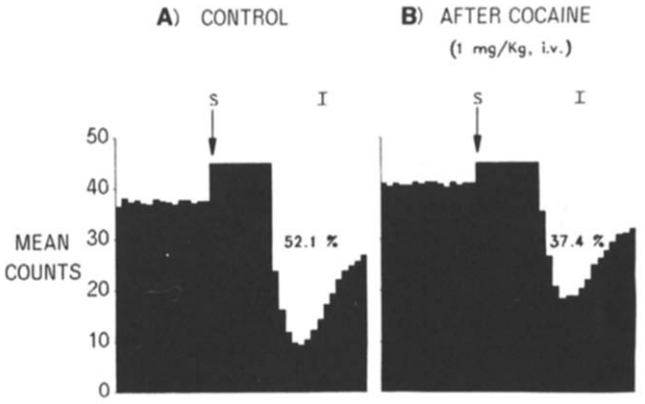

FIG. 12. Rate meter histograms showing the effects of cocaine on the inhibitory response of spontaneously active PC neurons to electrical stimulation of LC neurons. The left (control) panel represents a computer generated histogram of the mean counts per 1 second bin for 5 single PC neurons. Baseline activity is shown for a period of 15 seconds prior to the stimulus (S). The stimulus artifact is represented by the elevation in mean counts over baseline for 10 seconds. This histogram is constructed from 5 stimulation trials (conducted every two minutes) for each of the 5 neurons. The subsequent stimulus induced inhibition (I) is a mean value of 52.1%. After cocaine (1 mg/kg, IV) administration the same procedure was followed for the same 5 neurons. The post-drug stimulus induced inhibition is a mean value of 37.4%.

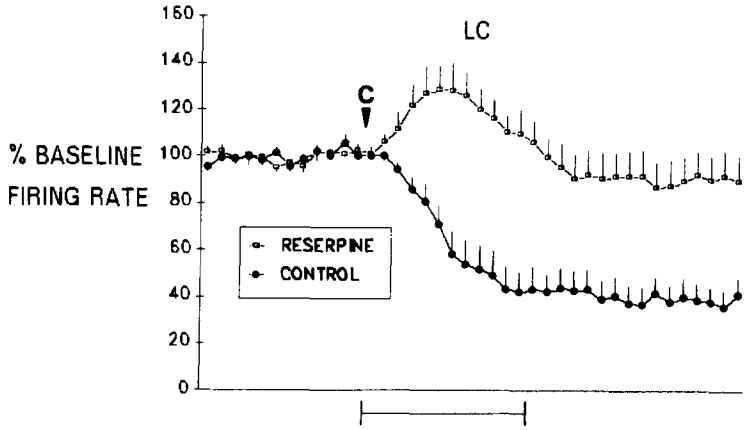

FIG. 13. Time course for the effects of cocaine on LC neuron firing rate in control (n=9) and reserpine pretreated animals (n=5; 10 mg/kg, IP; 5 hours before electrophysiological recording). Cocaine administration is indicated by an arrow (C, 1 mg/kg, IV). The calibration bar represents a period of 2 minutes. The data is depicted as mean percent of baseline firing rate, and is constructed from 10 second bins from individual neurons.

cocaine (1 mg/kg, IV) on LC neuron firing in reserpine treated (n=5) and control animals (n=9). In control animals, cocaine invariably elicited an inhibition of neuron firing. However, the predominant effect of cocaine in reserpine treated animals was an activation of neuron firing. The transient increase in activity in reserpinized animals was readily apparent one minute after cocaine administration, when 80% of the neurons were excited (122.6±4.5 percent of baseline) and only 10% of the neurons were inhibited (91.9 percent of baseline) and 10% showed no effect (n=10). Table 1 illustrates the transient nature of the excitatory response in the reserpinized animals where only 30% of the neurons were still excited two minutes after cocaine administration. The mean percent of baseline firing activity of reserpinized and control animals two minutes after cocaine administration were 102.1±6.3% (n=10) and 38.7±7.7% (n=12) respectively and were significantly different ($p<0.001$).

Figure 14 compares the time course for the effects of

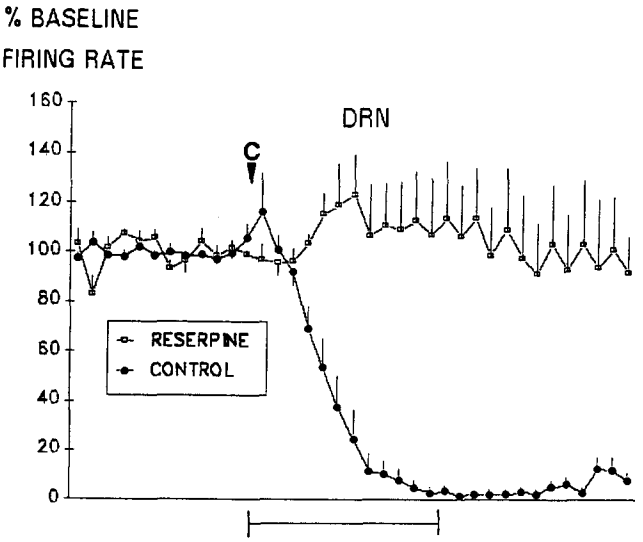

FIG. 14. Time course for the effects of cocaine on the firing rate of DRN neurons in control (n=6) and reserpine pretreated animals (n=5). See Fig. 13 for further details.

cocaine (1 mg/kg, IV) on DRN firing in reserpine treated (n=6) and control animals (n=6). In control animals, cocaine invariably elicited an inhibition of neuron firing. However, the predominant effect of cocaine in reserpine treated animals was an activation of neuron firing (75%, Table 1). The mean percent of baseline firing two minutes after cocaine administration was significantly different for the reserpinized and control animals (126.9±23.2%, n=8 and 3.0±3.0%, n=6 respectively; $p=0.001$, Mann-Whitney test).

DISCUSSION

One of the mechanisms accounting for the observed inhibition of central monoaminergic neurons by cocaine is most likely an inhibition of transmitter reuptake. Since all of the monoaminergic neurons studied were spontaneously firing and all are considered to employ a reuptake mechanism [10] for the recapture and inactivation of synaptically released transmitter, it follows that inhibition of reuptake of tonically released transmitter at neuron terminals, will potentiate the synaptic actions of the transmitter. The results obtained in reserpinized animals support the blockade of reuptake hypothesis. However, a few problems exist with the ability of this hypothesis to totally account for the central actions of cocaine: (1) cocaine antagonized rather than potentiated the inhibitory effects of LC stimulation on Purkinje neuron firing; (2) cocaine was equivocal in inhibiting dopaminergic neurons. Such data implies that mechanisms/hypotheses other than "blockade of reuptake" should also be considered. For the "inhibition of reuptake" hypothesis, the source of transmitter may be the axon collaterals [1] or dendritic synapses [15]. The synapses could arise from the monoaminergic neurons within the nucleus or monoaminergic inputs originating elsewhere but terminating in the pertinent area [18]. Electrophysiological or lesion studies cannot as yet unequivocally differentiate the contribution of one input relative to the others.

The inhibitory effect of cocaine on the monoaminergic neurons studied cannot be accounted for by its local anesthetic properties or cardiovascular effects for five reasons: (1) cocaine at the doses studied never decreased action potential amplitude or slope; (2) procaine, a structurally related local anesthetic which shared some of the cardiovascular effects of cocaine in conscious and barbiturate anesthetized animals, did not elicit any significant effects on the neurons studied; (3) the inhibitory effects of cocaine on locus coeruleus or dorsal raphe neurons in reserpinized animals were very significantly attenuated, and many cells were in fact initially excited; (4) the central inhibitory effects of cocaine were not directly correlated with the relatively brief increase in mean arterial pressure resulting from systemic administration; and (5) specific adrenoceptor antagonists (yohimbine, piperoxane, haloperidol) were able to antagonize or reverse the effects of cocaine on LC and VTA neurons.

The inhibitory effects of cocaine reported here provide an interesting comparison to the effects of amphetamine and tricyclic antidepressants on these neurons as reported previously. Amphetamine has been shown to *inhibit* central dopaminergic [7] and noradrenergic neurons [14], but in lesioned animals to *activate* central serotonergic neurons [4]. Tricyclic antidepressants have been reported to inhibit central noradrenergic neurons (2 tricyclics—2) and serotonergic neurons (3 tricyclics—2) and are generally thought to be devoid of effects on dopaminergic neurons [3]. Clearly, the present data with cocaine, along with previous studies with amphetamine and tricyclic antidepressants, highlight the similarities and differences between the effects of cocaine, amphetamine and tricyclic antidepressants on central monoaminergic neurotransmission.

Our results also suggest that the effects of cocaine on central monoaminergic neurotransmission are *not* directly due to its local anesthetic properties or its cardiovascular effects.

The potent inhibitory effects of cocaine on some central presynaptic monoaminergic neurons also implies that areas postsynaptic to such neurons would be significantly affected by cocaine—especially if such postsynaptic neurons are follower cells for tonically active presynaptic inputs. Obviously, the effects of cocaine on such regions and the monoaminergic neurons described would collectively contribute to its psychotropic and other centrally mediated effects.

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