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Dopaminergic Substrates of Cocaine-Induced Place Conditioning

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Recently, Spyraki et al. (*Brain Research*, 253 (1982) 195–203) reported that cocaine-induced place preference conditioning was unaffected by blockade of central dopamine (DA) or norepinephrine function. In addition, systemic injections of the local anesthetic procaine produced place preference conditioning. The present study was undertaken to further evaluate the possible role of DA in cocaine-induced place conditioning. In Expt. 1, a partial replication of Spyraki et al., systemic cocaine (5.0 mg/kg, i.p.) produced significant place conditioning that was not disrupted with the DA antagonist pimozide (1.0 mg/kg, i.p.). In Expt. 2, cocaine was microinjected unilaterally into the lateral ventricles to eliminate peripheral local anesthesia. Cocaine (50.0 μg, i.c.v.) produced place conditioning and pretreatment with pimozide (1.0 mg/kg, i.p.) disrupted the effect. In Expt. 3, place conditioning was not observed when cocaine presentations (50.0 μg, i.c.v.) were paired with both compartments. The substrates of cocaine-induced place conditioning were further investigated in Expt 4: Procaine (250 μg, i.c.v.) did not produce place conditioning whereas the DA agonist bromocriptine (50.0 μg, i.c.v.) did. Results suggest the involvement of central DA in cocaine-induced place conditioning.

INTRODUCTION

The conditioned place preference paradigm is commonly used to study the reinforcing properties of various stimuli. Spyraki et al.[26–29] recently investigated the dopaminergic substrates of place preference conditioning and the results of 3 studies supported a dopaminergic theory of reward. Thus, the integrity of the mesolimbic dopamine (DA) system was shown to be critical for heroin reward[29] and a reduction of central DA function attenuated conditioned place preference induced by food[26] and by (+)-amphetamine[28].

However, Spyraki et al. obtained unexpected results when they studied the reinforcing properties of cocaine. Systemic administrations of cocaine produced a dose-related preference for the distinctive environment that had been paired with the drug[27]. In this case, however, disruption of central DA function did not influence the place preference; pimozide, haloperidol and 6-hydroxydopamine (6-OHDA) lesions of the nucleus accumbens were all ineffective[27]. Destruction of central and peripheral norepinephrine (NE) systems with 6-OHDA were equally ineffective in attenuating this cocaine-induced conditioned place preference[27].

Cocaine, an indirect cathecholamine (CA) agonist, inhibits the reuptake of DA[12,25] and NE[17]. In addition to these stimulant effects, cocaine has prominent local anesthetic properties[1] that are often assumed to be unrelated to its reinforcing properties[14,15]. However, there is evidence that procaine and other local anesthetics, which do not share cocaine's potent central stimulant effects, will support intravenous (i.v.) self-administration[6,11,15,31]. These data suggest that the reinforcing properties of cocaine might not be entirely related to its DA action. Spyraki et al. suggested that perhaps local anesthetic effects produced by intraperitoneal (i.p.) injection of cocaine might somehow contribute to place prefer-

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ence conditioning[27]. In support of this hypothesis, procaine, injected i.p. in doses that did not affect DA uptake in the CNS, induced conditioned place preference[27]. Spyraki et al. further postulated that "...if it were possible to block selectively the local anesthetic properties of cocaine, then it is quite conceivable that the drug would continue to produce place preference conditioning through its facilitation of DA neurotransmission"[27]. Unfortunately, methods for selective in vivo blockade of local anesthesia are not available. However, if small doses of cocaine were injected intracerebrally, the local anesthetic effects may be minimized.

The following experiments were designed to assess the dopaminergic substrates of cocaine-induced place conditioning. It was hypothesized that administering cocaine intracerebrally would lead to place conditioning and that neuroleptics would attenuate this effect. This would provide further evidence for a critial role of DA in place conditioning.

MATERIALS AND METHODS

Subjects

Male Wistar rats (200–250 g) were obtained from Charles River Canada. Animals were initially housed in groups of 8–10 in wire cages (50 × 30 × 15 cm) for a period of at least two weeks. After cannulation and during the behavioral testing period, all rats were individually housed in smaller wire cages (25 × 15 × 15 cm). Animals were maintained on a 12 h light (06.00–18.00 h)/dark cycle with food and water available ad libitum in the home cages. Behavioral testing was performed during the dark cycle (18.00–23.00 h).

Surgery

Animals to be cannulated were food-deprived for the 24 h before surgery. At the time of surgery, rats were anesthetized with sodium pentobarbital (60 mg/kg, i.p.) and secured in a Kopf stereotaxic instrument with the incisor bar set 5.0 mm above the interaural line. A stainless steel guide cannula (20 gauge) was implanted into one of the lateral ventricles at the coordinates A 0.0, L 1.7 and V 2.5 (ref. 21) and anchored to the skull with 4 stainless jeweller's screws and dental acrylic cement. The cannulae were protected between central injections with an obturator pin and sealed with silicone. Cannulae were implanted into the left lateral ventricle of half the animals and into the right lateral ventricle of the other half.

Drugs

Cocaine hydrochloride (BDH Chemicals, Toronto) and procaine hydrochloride (Sigma Chemical Co.) were dissolved in saline. Bromocriptine mesylate (Sandoz Pharmaceuticals) was dissolved in 40% propylene glycol. Pimozide (Janssen Pharmaceutica) was dissolved in a ratio of 6 parts tartaric acid to 1 part pimozide (by weight) in boiling distilled water and cooled to room temperature prior to injection.

Apparatus

Four 3-compartment wooden shuttle boxes (25 × 32 × 89 cm) were outfitted with a Plexiglas floor and cover. Two large compartments (25 × 32 × 34 cm) were separated by two guillotine doors from a central area (11 × 25 × 32 cm). These two end compartments was distinguished by brightness, odor, and texture. The walls of these end compartments and the corresponding side of each guillotine door were painted white or black. The Plexiglas floor of the white compartment was covered with wood shavings. To contrast the slight wood odor, the black Plexiglas floor of the black compartment was dampened with a mild acetic acid solution (2.5%). The small central compartment, a neutral unpainted area, served as a choice point in Phases I and III of the experiment. The floors were washed and recovered with fresh wood shavings and acetic acid after every rat.

General experimental procedure

Behavioral testing was carried out over 12 consecutive days. During the initial preconditioning phase (Phase I), the animals were habituated to the apparatus for 15 min on 3 consecutive days. Each rat was placed in the middle compartment for 30 s, then the two guillotine doors were removed thereby allowing the rat to explore the 3 compartments of the shuttle box for 15 min. On the third day, the time spent by each animal in the two end compartments was manually recorded. An animal was considered to be in a compartment only when both front paws were in that particular compartment.

The conditioning phase (Phase II) consisted of 8 30-min sessions: 4 vehicle and 4 drug pairings. Ani-

mals were restricted to one end compartment by the guillotine door during the conditioning sessions. Rats injected with the drug were immediately placed in their initially (Phase I) less preferred side, whereas rats injected with the vehicle were placed in their initially preferred side (except in Expt 3, see below). Vehicle and drug pairings were administered on alternating days, with a counterbalanced order of presentation.

A postconditioning test was held on day 12 (Phase III). On this test day neither vehicle nor drug was administered. As in Phase I, the drug-free subjects were placed into the middle compartment for a 30-s waiting period and subsequently given access to the entire shuttle box. Again the time spent in each of the two distinctive compartments was manually recorded during the 15-min period. The recordings (Phases I and III) were performed by an experimenter who was blind to previous pairing conditions.

Intracerebroventricular injections

Manual intracerebroventricular (i.c.v.) microinjections of the drug or the vehicle were delivered in a volume of 1 μl by a 5 μl Hamilton microsyringe. Injection cannulae were constructed with 26-gauge stainless steel tubing, cut to extend 1.0 mm beyond the tip of the guide cannula, and attached to the microsyringe by a length of polyethylene tubing.

Animals were hand-restrained during the insertion of the injection cannula into the outer cannula. The rats were then placed in one of the two end compartments of the shuttle box and allowed to move freely for 2 min. At this point the 1 μl injection was delivered in 20–30 s and the injection cannula was left in place for an additional 60 s to ensure sufficient diffusion and avoid withdrawing drug during removal of the injection system. After the 30-min conditioning period, an obturator pin was reinserted into the guide cannula and secured with sealant.

Histological examiniations

At the conclusion of behavioral testing, animals were sacrificed for histological confirmation of cannula placements. Rats were injected with an overdose of sodium pentobarbital, exsanguinated with intracardial saline followed by 10% Formalin. Frozen coronal sections (40 μm) were taken through the lateral ventricles. The serial brain sections were floated in saline, mounted, and stained with formol-thionin[5]. Additional brain sections were mounted on microscope slides and placed on a photographic enlarger to produce prints of unstained sections.

EXPERIMENT 1 — SYSTEMIC ADMINISTRATION OF COCAINE

In a partial replication of Spyraki et al.[27], cocaine was administered systemically (5.0 mg/kg, i.p.) to two groups ($n = 8$) of unoperated rats. These animals received the 3 phases described above except that one group was administered a systemic injection of pimozide (1.0 mg/kg, i.p.) 4 h prior to every conditioning session (Phase II); this dose was previously used by Spyraki et al.[27].

Results and Discussion

Time spent in the two end compartments of the shuttle box during the pre- and postconditioning test days was expressed as percent of time spent on the originally less preferred side (Phase I). Place conditioning is indicated by a relative increase in this percent during the postconditioning test session (Phase III). As shown in the top panel of Fig. 1, cocaine appeared to produce place conditioning that was not blocked by pimozide.

A two-factor analysis of variance (ANOVA) with one repeated measure was conducted to assess the significance of these effects. The two variables analyzed were phase (pre- and postconditioning) and group (cocaine and pimozide + cocaine). The analysis revealed a highly significant phase effect, $F_{1,14} = 14.76$, $P < 0.002$. The main effect of group was not statistically reliable, $F_{1,14} = 0.51$, $P > 0.50$, nor was the phase by group interaction, $F_{1,14} = 0.11$, $P > 0.70$. The non-significant interaction between the two variables suggests that the two drug treatments produced the same effect across the two phases.

Both groups showed place conditioning, i.e., a significant increase in the mean percent of time spent in the compartment that previously had been associated with cocaine. These findings are consistent with those of Spyraki et al.[27]; systemic cocaine (5.0 mg/kg) induced place conditioning which was not disrupted in rats pretreated with pimozide (1.0 mg/kg, i.p.). This might suggest that a non-dopaminergic mecha-

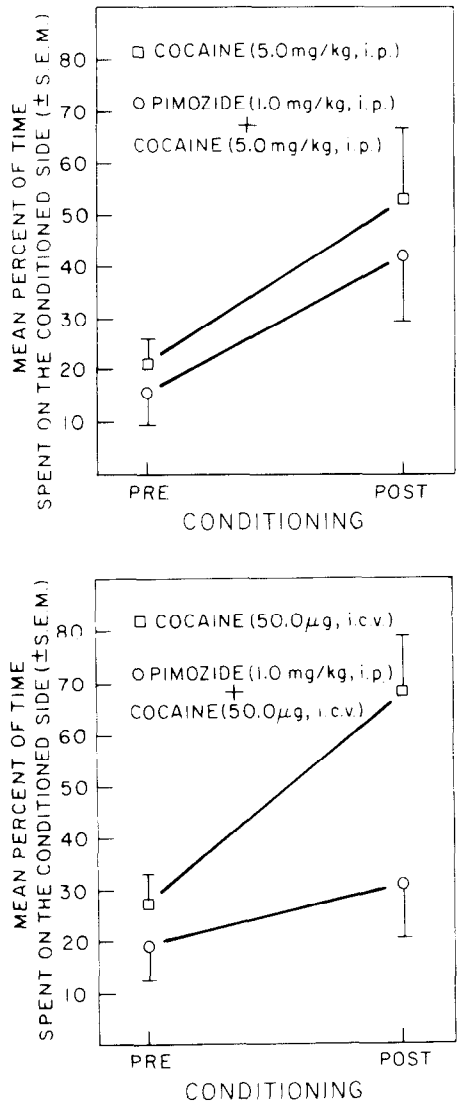

Fig. 1. Mean (± S.E.M.) percent of time spent on the conditioned side by the cocaine and pimozide + cocaine groups during the pre- and postconditioning test sessions of Expts. 1 (top panel) and 2 (bottom panel).

nism underlies the place conditioning. Possibly, the local anesthetic properties of cocaine contributed to this effect.

EXPERIMENT 2 — I.C.V. ADMINISTRATION OF COCAINE

Although methods to selectively block the local anesthetic properties of cocaine are not available, it was reasoned that microinjecting small quantities directly into the CNS would minimize the local anesthesia. To explore this possibility, the first experiment was repeated with cocaine and saline administered into the lateral ventricles of groups either pretreated or not pretreated with pimozide.

Twenty animals were stereotaxically implanted with chronic indwelling guide cannulae and randomly assigned to two groups (n = 10). These rats received the same 3 phases as in the first experiment except that cocaine (50.0 μg) or saline (1.0 μl) was injected i.c.v. prior to each conditioning session (Phase II). As in the previous experiment, one group received pimozide (1.0 mg/kg, i.p.) 4 h prior to each conditioning session.

Results and Discussion

Of the 20 operated rats, one animal (cocaine group) had a cannula placement which clearly did not penetrate the lateral ventricle. In addition, 3 animals were discarded because of defective cannulae. Therefore, 16 rats (n = 8 for both groups) with cannula placements directly into the lateral ventricle (Fig. 2) were included in the statistical analysis.

The data are presented and analyzed in the same manner as in Expt. 1. The lower panel of Fig. 1 illustrates the mean percent of time spent on the conditioned side during the pre- and postconditioning test sessions. The group receiving i.c.v. cocaine appeared to show place conditioning and the effect was attenuated in rats pretreated with pimozide. A two-factor ANOVA revealed a highly significant phase effect, $F_{1,14} = 16.11$, $P < 0.002$, an effect of group, $F_{1,14} = 4.76$, $P < 0.05$, and a phase by group interaction, $F_{1,14} = 4.86$, $P < 0.05$. Tests of simple main effects performed on the phase variable for the two groups revealed a phase effect in the cocaine group, $F_{1,14} = 19.33$, $P < 0.001$, but no significant phase effect in the pimozide + cocaine group, $F_{1,14} = 1.64$, $P > 0.10$.

The observation that i.c.v. cocaine produced place conditioning that was significantly disrupted by pimozide suggests that the local anesthetic effects did not contribute significantly to the conditioning. These findings support the view that place conditioning following cocaine was mediated by central DA-containing neurons.

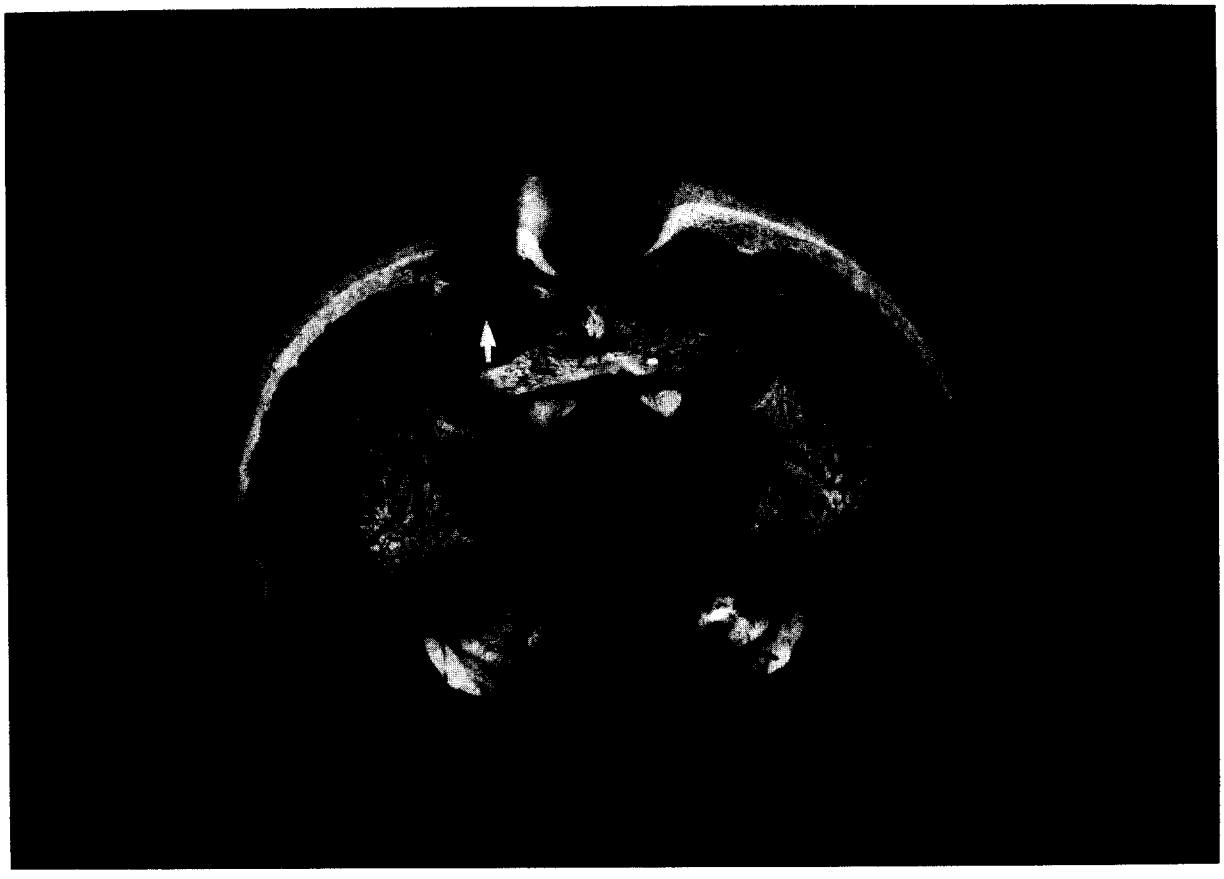

Fig. 2. Representative photomicrograph of an unstained brain section illustrating unilateral cannula tract penetrating the right lateral ventricle of the rat. The estimated location of the cannula tip is indicated by a white arrow.

EXPERIMENT 3 - COCAINE PAIRED WITH BOTH COMPARTMENTS

It is possible that a differential degree of locomotor activity during the two test sessions (Phases I and III) could have resulted in the differences in the percent of time spent on the conditioned side in Expts. 1 and 2. Thus, rats might have been more active in the second test session (Phase III) because of previous pairings of cocaine with the apparatus. This conditioning of general motor activity could have produced an increased number of crossings into the initially less preferred compartment and the resultant increase in time spent there. This alternative hypothesis is supported by a number of studies which have demonstrated that enhanced DA activity, produced by treatment with psychomotor stimulants, can lead to environment-specific conditioned locomotor activity[2]. For example, animals injected with cocaine and placed in a test environment on a series of daily sessions are observed to be more active when injected with saline and placed there[3,13,22].

The purpose of Expt. 3 was to assess this possibility. Animals received to cocaine pairings and two vehicle pairings in each compartment. A significant increase in mean percent of time spent on the 'conditioned' or initially less preferred side might reflect a general motor activation effect. In contrast, the lack of such a change would support the involvement of central DA in cocaine-induced place conditioning.

Nine rats were implanted with chronic indwelling cannulae. These animals received the same 3 phases described above except that drug and vehicle presentations were paired with both end compartments. Each rat received 2 cocaine pairings and 2 saline pairings in the black and in the white compartment. As in the previous experiments, drug and vehicle were presented on alternate days.

Results and Discussion

Eight animals were included in the statistical analysis; one rat was discarded because of a defective guide cannula. The mean percent of time spent on the 'conditioned' or initially less preferred end compartment during the pre- and postconditioning sessions is depicted in Fig. 3. There appeared to be little change across phases and a one-factor, repeated measures ANOVA revealed that the percent of time spent in the 'conditioned' side during the two test sessions was not significantly different, $F_{1,7} = 0.25$, $P > 0.63$.

Since locomotor activity was not recorded, it was not possible to determine if the animals exhibited environment-specific conditioned locomotor activity or an increased number of crossing between compartments. However, the results suggest that conditioned activity effects did not produce the results seen in the first two experiments.

EXPERIMENT 4 — ASSESSMENT OF DOPAMINERGIC AND LOCAL ANESTHETIC PROPERTIES

Dopaminergic substrates have been demonstrated for place conditioning induced by systemic amphetamine[28], apomorphine[30], food [26] and heroin[29]. A peripheral local anesthetic substrate has also been demonstrated[27]. To examine the individual contribution of these two substrates in the CNS, Expt. 4 assessed place conditioning following i.c.v. administrations of the DA agonist bromocriptine or the local anesthetic procaine. Bromocriptine is a DA agonist throught to be specific for the D_2 receptors[7]. Procaine was chosen as the local anesthetic because it induced place preference conditioning when administered systemically[27]. The dose of procaine used in this experiment was larger than the dose of cocaine because it is a less potent local anesthetic agent than cocaine[10].

Eighteen rats were implanted with chronic indwelling cannulae. These animals received the same 3 phases described in Expt. 2 but received bromocriptine (50.0 μg, i.c.v.) or procaine (250.0 μg, i.c.v.).

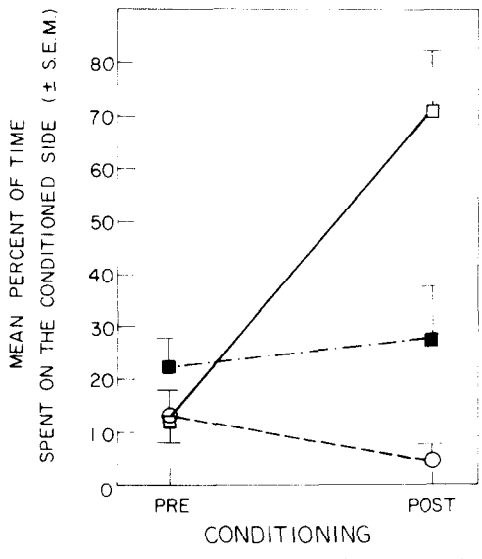

Fig. 3. Mean (± S.E.M.) percent of time spent in the 'conditioned' side during the pre- and postconditioning test sessions by rats that received cocaine (50.0 μg, i.c.v.) and vehicle presentations paired with both compartments of the shuttle box (■---■), bromocriptine (50.0 μg, i.c.v.) presentations (□——□) or procaine (250.0 μg, i.c.v.) presentations (○---○) during the conditioning phase of Expts. 3 and 4.

Results and Discussion

Of the 18 operated rats, 3 animals were discarded because of defective cannulae. In addition, histology revealed that 1 rat had a cannula placement which clearly did not penetrate the lateral ventricle. Therefore, 14 rats ($n = 7$ for both groups) with cannula placements directly into the lateral ventricle were included in the statistical analyses.

The mean percent of time spent in the compartment paired with i.c.v. bromocriptine before and after conditioning are illustrated in Fig. 3. The group showed a large increase in time spent on the conditioned side from pre-exposure to test. A one-way, within-subjects ANOVA revealed that this effect was significant, $F_{1,6} = 28.05$, $P < 0.003$.

In contrast, i.c.v. procaine did not induce place conditioning. Indeed, as depicted in Fig. 3, animals appeared to spend less time in the compartment that was paired with procaine and this approached significance, $F_{1,6} = 5.04$, $P < 0.07$. Thus, it appears that the DA agonist bromocriptine, but not the local anesthetic procaine, can induce place conditioning when administered i.c.v.

GENERAL DISCUSSION

The present study investigated the dopaminergic

substrates of cocaine-induced place conditioning. The hypothesis of Spyraki et al.[27] that cocaine's local anesthetic properties may be sufficient to produce place conditioning was supported by the results of the first experiment. Disruption of DA function with pimozide did not significantly influence place conditioning established by systemic cocaine.

Spyraki et al. suggested that "...if it were possible to block selectively the local anesthetic properties of cocaine, then it is quite conceivable that the drug would continue to produce place preference conditioning through its facilitation of DA neurotransmission"[27]. Although no attempts were made to selectively block the local anesthetic effects of cocaine in the present study, cocaine was administered intracerebrally to minimize peripheral local anesthesia. This produced place conditioning that was disrupted by the DA receptor blocker pimozide. Thus, cocaine did not appear to produce rewarding local anesthesia to a significant extent when administered in the lateral ventricle.

The individual contributions of the central local anesthetic and dopaminergic substrates to place conditioning were further assessed with pharmacologically specific agents. Procaine, a local anesthetic, did not produce place conditioning when administered i.c.v. However, pairing a compartment of the shuttle box with presentations of i.c.v. bromocriptine, a DA agonist, did result in significant place conditioning. These results support the notion that administering small quantities of cocaine directly into the CNS appears to be a viable method to assess the DA substrates of cocaine-induced place conditioning.

To investigate the possibility that cocaine-induced environment-specific conditioned locomotor activity could have inflated the amount of time spent in the initially less preferred compartment during the postconditioning test session, an additional experiment was conducted in which cocaine and vehicle presentations were paired with both compartments. Place conditioning was not observed in these animals, thereby suggesting that the effects observed in the other experiments were not simply the result of general motor activation. This control study is especially important in light of a recent report of an apparent conditioned place preference obtained when morphine was paired with both sides of the shuttle box[20]. The discrepency between these two similar control studies could arise from differences in drug or any of the several methodological differences.

A role for central DA systems in place conditioning induced by i.c.v. cocaine is consistent with evidence from the self-administration paradigm. Neuroleptics and 6-OHDA can attenuate intravenous[4,23,24] and intracerebral[8,9] cocaine self-administration. Yet, these treatments fail to block place conditioning induced by i.p. cocaine[27]. There are several possible explanations for this discrepancy. For example, the place conditioning induced by i.p. cocaine and procaine was probably induced by peripheral local anesthesia. Assuming that there is some level of pain or discomfort associated with an i.p. injection, the local anesthetic properties of cocaine and procaine would appease it whereas this level of pain might linger after a vehicle injection. This differential level of pain associated with the two distinctive compartments could have resulted in the increased amount of time spent in the originally less preferred compartment (the one paired with cocaine or procaine injections) during the postconditioning session. This problem would appear to have been avoided by i.c.v. administrations[19].

Others have found that neuroleptics and 6-OHDA lesions failed to block place conditioning with two other indirect CA agonists, methylphenidate (MPD) and nomifensine (NFS)[18]. Since neither share cocaine's local anesthetic properties, Martin-Iverson et al. proposed that "...the failure to disrupt conditioned place preference could be a general property of non-amphetamine psychostimulants" and that the place preferences induced by these agents were mediated by "...presently unspecified non-catecholaminergic neural substrates"[18]. However, the highest dose of haloperidol (1.0 mg/kg) did attenuate the place conditioning induced by MPD and the effects of this dose on NFS-induced place conditioning were not evaluated[18]. In addition, the DA depletions induced by 6-OHDA lesions never exceeded 85% (ref. 18) and it has been suggested that extensive lesions ($> 90\%$) are required to reduce the rewarding properties of psychostimulants[23,28]. In view of these problems, a second explanation was postulated to explain their results. The authors proposed that "...even a slight increase in activation of DA receptors may be sufficient to produce a rewarding effect"[18]. The present demonstration of a dopaminergic substrate of co-

caine-induced place conditioning would suggest that a careful re-evaluation of this second explanation should be given to place conditioning induced by MPD and NFS.

In conclusion, the evidence obtained in the present study supports the involvement of central DA in cocaine-induced place conditioning. In conjunction with evidence from the self-administration paradigm, these results support the involvement of central DA systems in cocaine reward.

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REFERENCES

1 Bedford, J.A., Turner, C.E. and Elsohly, H.N., Local anesthetic effects of cocaine and several extracts of the coca leaf (*E. coca*), *Pharmacol. Biochem. Behav.*, 20 (1984) 819–821.
2 Beninger, R.J., The role of dopamine in locomotor activity and learning, *Brain Res. Rev.*, 68 (1983) 173–196.
3 Beninger, R.J. and Herz, R.S., Pimozide blocks establishment but not expression of cocaine-produced environment-specific conditioning, *Life Sci.*, 38 (1986) 1425–1431.
4 De Wit, H. and Wise, R.A., Blockade of cocaine reinforcement in rats with the dopamine receptor blocker pimozide, but not with the noradrenergic blockers phentolamine and phenoxybenzamine, *Can. J. Psychol.*, 31 (1977) 195–203.
5 Donovick, P., A metachromatic stain for neural tissue, *Stain Technol.*, 49 (1974) 49–51.
6 Ford, R.D. and Balster, R.L., Reinforcing properties of intravenous procaine in rhesus monkeys, *Pharmacol. Biochem. Behav.*, 6 (1977) 289–296.
7 Fuxe, K., Ogren, S.O., Agnati, L.F., Andersson, K., Hall, H., Kohler, C. and Fredholm, B., Central monoaminergic synapses as sites of action for ergot drugs. In M. Goldstein, D.B. Calne, A. Lieberman and M.O. Thorner (Eds.), *Ergots Compounds and Brain Function, Adv. Biochem. Psychopharmacol. Vol. 23*, Raven Press, New York, 1980, pp. 41–62.
8 Goeders, N.E. and Smith, J.E., Cortical dopaminergic involvement in cocaine reinforcement, *Science*, 221 (1983) 773–775.
9 Goeders, N.E. and Smith, J.E., Effects of 6-hydroxydopamine lesions of the medial prefrontal cortex on intracranial cocaine self-administration. *Soc. Neurosci. Abstr.*, 10 (1984) 1206.
10 Goldstein, A., Aronow, L. and Kalman, S.M., *Principles of Drug Action: The Basis of Pharmacology*, 2nd edn., Wiley, New York, 1974, 854 pp.
11 Hammerbeck, D.M. and Mitchell, C.L., The reinforcing properties of procaine and D-amphetamine compared in rhesus monkeys, *J. Pharmacol. Exp. Ther.*, 204 (1978) 558–569.
12 Heikkila, R.E., Cabbat, F.S., Manzino, L. and Duvoisin, R.C., Rotational behavior induced by cocaine analogs in rats with unilateral 6-hydroxydopamine lesions of the substantia nigra: dependence upon dopamine uptake inhibition, *J. Pharmacol. Exp. Ther.*, 211 (1979) 189–194.
13 Hinson, R.E. and Poulos, C.X., Sensitization to the behavioral effects of cocaine: modulation by pavlovian conditioning, *Pharmacol. Biochem. Behav.*, 15 (1981) 559–562.
14 Jaffe, J.H., Drug addiction and drug abuse. In A. Goodman Gilman, L.S. Goodman and A. Gilman (Eds.), *The Pharmacological Basis of Therapeutics*, 6th edn., MacMillan, Toronto, 1980, pp. 535–584.
15 Johanson, C.E., The reinforcing properties of procaine, chloroprocaine and proparacaine in rhesus monkeys, *Psychopharmacology*, 67 (1980) 189–194.
16 Keppel, G., *Design and Analysis A Researcher's Handbook*, 2nd edn., Prentice-Hall, New Yersey, 429 pp.
17 Langer, S.Z. and Enero, M.A., The potentiation of responses to adrenergic nerve stimulation in the presence of cocaine: its relationship to the metabolic fate of released norepinephrine, *J. Pharmacol. Exp. Ther.*, 191 (1974) 431–443.
18 Martin-Iverson, M.T., Ortmann, R. and Fibiger, H.C., Place preference conditioning with methylphenidate and nomifensine, *Brain Research*, 332 (1985) 59–67.
19 Morency, M.A. and Beninger, R.J., Conditioned place preference induced by intracerebroventricular microinjections of cocaine: possible involvement of central dopamine. *Soc. Neurosci. Abstr.*, 11 (1985) 718.
20 Mucha, R.F. and Iverson, S.D., Reinforcing properties of morphine and naloxone revealed by conditioned place preferences: a procedural examination, *Psychopharmacology*, 82 (1984) 241–247.
21 Pellegrino, L.J., Pellegrino, A.S. and Cushmann, J.A., *A Stereotaxic Atlas of the Rat Brain*, 2nd edn., Plenum, New York, 1979.
22 Post, R.M., Lockfeld, A., Squillace, K.M. and Contel, N.R., Drug-environment interaction: context dependency of cocaine-induced behavioral sensitization, *Life Sci.*, 28 (1980) 755–760.
23 Roberts, D.C.S., Corcoran, M.E. and Fibiger, H.C., On the role of ascending catecholamine systems in intravenous self-administration of cocaine, *Pharmacol. Biochem. Behav.*, 6, 615–620.
24 Roberts, D.C.S., Koob, G.F., Klonoff, P. and Fibiger, H.C., Extinction and recovery of cocaine self-administration following 6-hydroxydopamine lesions of the nucleus accumbens, *Pharmacol. Biochem. Behav.*, 12 (1980) 781–787.
25 Snyder, S.H. and Coyle, J.T., Regional differences in ^{3}H-dopamine uptake into rat brain homogenate, *J. Pharmacol. Exp. Ther.*, 165 (1969) 78–86.

26 Spyraki, C., Fibiger, H.C. and Phillips, A.G., Attenuation by haloperidol of place preference conditioning using food reinforcement, *Psychopharmacology,* 77 (1982) 379–382.
27 Spyraki, C., Fibiger, H.C. and Phillips, A.G., Cocaine-induced place preference conditioning: lack of effects of neuroleptics and 6-hydroxydopamine lesions, *Brain Research,* 253 (1982) 195–203.
28 Spyraki, C., Fibiger, H.C. and Phillips, A.G., Dopaminergic substrates of amphetamine-induced place preference conditioning, *Brain Research,* 253 (1982) 185–193.
29 Spyraki, C., Fibiger, H.C. and Phillips, A.G., Attenuation of heroin reward in rats by disruption of the mesolimbic dopamine system, *Psychopharmacology,* 79 (1983) 278–283.
30 Van der Kooy, D., Swerdlow, N.R. and Koob, G.F., Paradoxical reinforcing properties of apomorphine: effects of nucleus accumbens and area postrema lesions, *Brain Research,* 259 (1983) 111–118.
31 Woolverton, W.L. and Balster, R.L., Reinforcing properties of some local anesthetics in rhesus monkeys, *Pharmacol. Biochem. Behav.,* 11 (1979) 669–672.