Brain Research, 327 (1985) 143–151
Elsevier

BRE 10525

Involvement of Nigrostriatal Dopamine Neurons in the Contraversive Rotational Behavior Evoked by Electrical Stimulation of the Lateral Hypothalamus*

EDWARD CASTAÑEDA, TERRY E. ROBINSON and JILL B. BECKER

Psychology Department and Neuroscience Laboratory Building, The University of Michigan, 1103 E. Huron St., Ann Arbor, MI 48104-1687 (U.S.A.)

(Accepted May 15th, 1984)

Key words: dopamine — electrical stimulation — rotational behavior — lateral hypothalamus — striatum — substantia nigra — 6-hydroxydopamine

This experiment was conducted to determine if nigrostriatal dopamine (DA) neurons are necessary for the contraversive rotational behavior evoked by electrical stimulation in the lateral hypothalamus. Rats were tested daily for electrical stimulation-induced rotational behavior (ESRB) for 5 days, and then given an injection of 6-hydroxydopamine (6-OHDA) or saline into the ipsilateral substantia nigra. The nearly total depletion of striatal DA (> 96%) completely abolished contraversive ESRB and resulted in the appearance of ipsiversive ESRB. Partial DA depletion (< 95%) had no effect on contraversive ESRB. In animals with a partial DA depletion subsequent treatment with a low dose of α-methyl-*p*-tyrosine (40 mg/kg) attenuated contraversive ESRB, while having no effect on control animals, or the ipsiversive turning in animals with > 96% DA depletion. We conclude that the nigrostriatal DA system is necessary for contraversive rotational behavior evoked by lateral hypothalamic stimulation, but that only a small percentage of DA fibers are required to maintain apparently 'normal' function — at least as indicated by contraversive ESRB.

INTRODUCTION

It is widely believed that the rotational (turning) behavior produced by dopamine (DA)-mimetic drugs[23,36,37] is due to an asymmetry in the functional activity of nigrostriatal DA neurons, and that this asymmetry causes animals to turn away from the more active side (for reviews see refs. 20, 21 and 29). Most of the evidence for a causal relationship between an asymmetry in the functional activity of nigrostriatal DA neurons and rotational behavior is from indirect pharmacological experiments. Relatively more direct evidence is provided by studies showing that electrical stimulation of nigrostriatal cells, or their fibers coursing through the lateral hypothalamus (LH), evokes rotational behavior away from the stimulated side (contraversive)[2,4]. This contraversive electrical stimulation-induced rotational behavior (ESRB) is thought to be dependent on DA-containing cells because stimulation at effective sites releases DA in the striatum[3,28], and because contraversive ESRB is attenuated by systemically administered DA antagonists[5,31,39].

However, the hypothesis that specifically DA neurons projecting from the substantia nigra to striatum are necessary for contraversive ESRB elicited by LH stimulation has never been directly tested. In fact, Saranak and Goldfarb[32,33], recently suggested that nigrostriatal DA neurons are *not* necessary for ESRB, because the depletion of striatal DA produced by intrahypothalamic 6-hydroxydopamine does not attenuate the contraversive ESRB elicited by stimulation of the substantia nigra. Unfortunately, it is difficult to directly compare the earlier studies by Arbuthnott and colleagues with those of Saranak and Goldfarb, because in most of the former experiments rotational behavior was evoked by stimulation in the LH; not the substantia nigra. It is possible that the contraversive ESRB elicited by stimulation in the LH has a different neural basis than that

* An abstract of this research was published previously in *Soc. Neurosci. Abstr.*, 81 (1982) 360.

Correspondence: T. E. Robinson, Neuroscience Laboratory Building, The University of Michigan, 1103 E. Huron St., Ann Arbor, MI 48104-1687, U.S.A.

elicited by nigral stimulation.

Therefore, the purpose of the present experiment was to determine if nigrostriatal DA neurons are necessary for the contraversive ESRB elicited by electrical stimulation in or near DA fibers coursing through the LH. We report they are.

MATERIALS AND METHODS

Subjects

Thirty-one adult male Holtzman rats (Madison, WI) weighing 300–400 g were housed individually on a reversed 14:10 h, light:dark cycle (lights off at 09.00 h) with food and water freely available.

Surgical procedures

Each rat was anesthetized with Equithesin, and a bipolar stainless steel, teflon-coated stimulating electrode (150 μm diameter, Plastic Products Co., MS303/3) was placed stereotaxically into the nigrostriatal bundle as it courses through the posterior-lateral hypothalamus (coordinates, 3.0–3.5 mm posterior to bregma, 1.8 mm to the right of the sagittal suture and 8.4 mm ventral from the skull surface; bregma and lambda on the same horizontal plane). In addition, a 22-gauge stainless steel guide cannula was placed above the ipsilateral rostral substantia nigra (coordinates, P 5.0, L 2.0 and V 5.0 mm).

Procedures for behavioral testing

The electrical stimulus was provided by a Grass S8 stimulator connected in series with a Grass constant current unit, and consisted of monophasic rectangular pulses of 0.1 ms duration presented at a rate of 50 pulses/s. During each test session the animals were placed in an 18.5 × 25 × 45 cm high Plexiglas observation cage, the stimulating electrodes connected, and 10 min allowed for habituation. Each animal then received 5 10 s stimulation trials during each daily test session with the following stimulus intensities, and in the following order: 100, 150, 200, 250, and 300 μA. There were 2 min between each trial.

Following 1 week of recovery from surgery all the animals were screened twice for ESRB, as described above. Only those animals that showed consistent turning contralateral to the stimulating electrode (contraversive) were used in the subsequent experiments (n = 21). In the remaining animals (n = 10) stimulation produced various forced behaviors, jumping, rearing or general hyperactivity, but not rotational behavior. Two weeks after surgery those animals that showed contraversive ESRB were tested daily for 5 days, as described above. On each day of baseline testing the number of one-quarter turns (90°) made to the left or right during each 10 s trial was recorded. Immediately following the 5th day of baseline testing each rat received an i.p. injection of 25 mg/kg of desipramine[7]. Thirty min later the rats were lightly anesthetized with ether and a 28-gauge stainless steel injection cannula was lowered through the chronically implanted guide cannula into the rostral zona compacta of the substantia nigra. Fifteen of the rats received an infusion (1.6 μl/min) of 6-hydroxydopamine hydrobromide (6-OHDA; 8 μg in 4 μl) and 6 rats 4 μl of the vehicle (0.9% saline). Daily testing for ESRB resumed the next day for an additional 8 days (post-treatment days 1–8). Immediately following behavioral testing on the 8th day all rats received an i.p. injection of α-methyl-*p*-tyrosine (α-MPT; 40 mg/kg, suspended in Tween 20) to further deplete brain DA[34]. At 2, 4 and 6 h after α-MPT treatment each rat was again tested for ESRB at each of the 5 current levels.

Neurochemical methods and procedures

Following all behavioral testing the animals were decapitated. The striatum, olfactory tubercle, and nucleus accumbens were rapidly dissected on ice, weighed, and placed into tubes containing 400 μl of 0.05 N $HClO_4$, with dihydroxybenzylamine (2.5 ng/10 μl) added as an internal standard. The tissue samples were homogenized and centrifuged at 5000 *g* for 45 min at 2–4 °C. Cerebellar tissue was homogenized and the homogenate was spiked with known concentrations of catecholamine (CA) standards. These standards were centrifuged and processed with the tissue homogenates. CA were then purified by adsorption onto alumina (adapted from ref. 16). Briefly, the supernatant was transferred to conical tubes containing 20 mg of acid-washed alumina in 200 μl 3 M Tris buffer (pH 8.6); vortexed for 1 min and reciprocally shaken for 10 min. The samples were then washed in 600 μl of 6 mM Tris (pH 8.6), followed by two consecutive washings with 600 μl of high purity distilled water. After brief centrifugation, the supernatant was discarded and 200 μl 0.05 N

$HClO_4$ added to the alumina to extract the CA. After a 1 min vortex, and brief centrifugation to remove fine alumina particles, the supernatant containing the CA was placed in vials and stored at −20 °C.

CA concentrations were measured by high performance liquid chromatography with electrochemical detection (HPLC-EC) utilizing a Brownlee RP-18, 5 μm, C-18, reverse-phase column. The electrochemical detectors (LC-4 and LC-4A, Bioanalytical Systems) employed glassy carbon electrodes set at +0.74 V against Ag/AgCl reference electrodes. The mobile phase was a 0.1 M citrate/phosphate buffer containing 0.1 mM EDTA, octyl sulfate, and methanol, with an apparent pH 3.35. Octyl sulfate concentration varied from 10–400 mg/ml and methanol from 5–15% depending on the column and its age. Under these conditions 20 μl of the sample was injected onto the column via a Rheodyne injection valve.

Four control and 4 rats with 6-OHDA lesions were killed upon completion of behavioral testing on the 8th post-treatment day, 6 h after α-MPT treatment. The remaining experimental animals were killed at least 5 days after treatment with α-MPT. To determine the effect of α-MPT on DA levels in normal animals, experimentally naive rats received i.p. injections of either 40 mg/kg of α-MPT (n = 7) or an equal volume of Tween 20 (n = 3) and were killed 6 h later. Tissue concentrations of DA were determined as described above.

RESULTS

Baseline testing

During the 5 day period of baseline testing electrical stimulation elicited consistent and reliable contraversive circling in all animals. As the stimulus intensity increased there was a linear increase in the rate of turning (r = +0.985), over the range of current intensities tested.

Effects of intra-nigral 6-OHDA on rotational behavior

Immediately after testing on the 5th day of baseline 15 rats received intra-nigral 6-OHDA and 6 an equal volume of saline. Testing for ESRB continued the next day for an additional 8 days. Fig. 1A shows the average baseline rate of turning, and the average number of rotations on each of the subsequent 8 days, for the control and experimental (6-OHDA) groups tested at 300 μA. 'Baseline' consists of the average number of rotations over all 5 days of baseline testing.

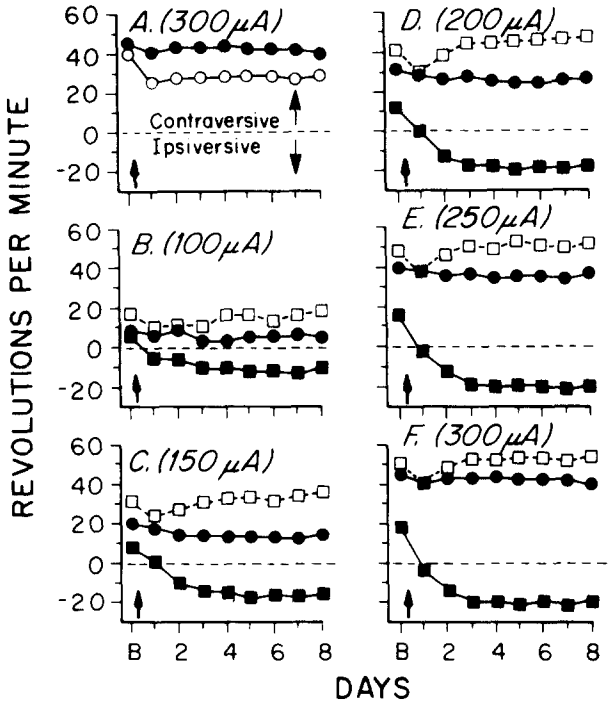

Fig. 1. Effects of intranigral 6-OHDA on rotational behavior evoked by electrical stimulation of the lateral hypothalamus. Each panel (A–F) gives the average number of rotations (expressed as revolutions/min) for different groups during 'baseline' (B), and for 8 days (days 1–8) after 6-OHDA or saline infusion into the substantia nigra (indicated by the vertical arrow). 'Baseline' consists of the average number of turns made over all 5 days of baseline testing. The current intensity used is noted in each panel. Contraversive rotations are expressed as positive numbers and ipsiversive rotations as negative numbers. Panel A: turning rates for control (closed circles) and all the experimental (6-OHDA injected; open circles) animals tested at 300 μA. There were no statistical differences between these two groups at this or any other current intensity when all experimental animals were grouped together. Comparisons made with profile analyses (see text). Panels B–F: turning rates for controls (closed circles), NONSWITCHERS (open squares, dashed line), and SWITCHERS (closed squares). Control animals did not differ from NONSWITCHERS at any current intensity. SWITCHERS differed from both controls and NONSWITCHERS at all current intensities. *P*-values of comparisons between SWITCHERS vs controls and NONSWITCHERS, respectively: Panel B, 100 μA, $P < 0.001$ and 0.047; C, 150 μA, $P < 0.02$ and 0.011; D, 200 μA, $P < 0.001$ and 0.002; E, 250 μA, $P < 0.001$ and 0.035; F, 300 μA, $P < 0.001$ and 0.001.

It would appear from examination of Fig. 1A (the

pattern was the same for all other current intensities) that the 6-OHDA infusion had little or no effect on contraversive ESRB ($P > 0.1$ for all current intensities). However, a closer examination of individual animals revealed that the 6-OHDA infusion resulted in two *qualitatively* different effects (Fig. 1B–F). In the majority of rats (n = 10) there was a transient, but non-significant, decline in contraversive ESRB on the day following 6-OHDA, followed by full recovery to baseline levels within 1 day (Fig. 1B–F). There was actually a non-significant tendency for these animals to show elevated levels of contraversive ESRB by 2–3 days following 6-OHDA (c.f. ref. 32). For the purpose of brevity we will call these animals 'NONSWITCHERS'. In the remaining one-third of the animals 6-OHDA resulted in a dramatic and total cessation of contraversive ESRB within 2–3 days, coincident with the development of relatively vigorous *ipsiversive* ESRB. Over 3–4 days these animals, which we will call 'SWITCHERS', increased their rate of ipsiversive ESRB and thereafter showed stable rates of rotation — but in the opposite direction to baseline (Fig. 1B–F).

Fig. 1B–F illustrates the effects of 6-OHDA on rotational behavior when SWITCHERS and NONSWITCHERS are examined separately. We think this post-hoc analysis is justified for two major reasons: (1) the 6-OHDA infusion produced two *qualitatively* different effects (i.e., switching or not switching direction); and (2) the analysis of striatal DA concentrations (presented below) revealed there was no overlap between the SWITCHERS and NONSWITCHERS in the extent of the DA depletion (i.e., neurochemically these are two distinct groups). To statistically compare SWITCHERS, NONSWITCHERS and controls, contraversive rotations were arbitrarily assigned positive values and ipsiversive rotations negative values (see Fig. 1B–F). The groups were then compared using profile analyses[25], conducted separately for each current intensity. There were no statistical differences between control animals and NONSWITCHERS at any current intensity. In contrast, the SWITCHERS differed significantly from both the controls and NONSWITCHERS at all current intensities (Fig. 1B–F; F values ranged from 4.8 to 50.4, $P < 0.047 - < 0.001$). Separate analyses of baseline turning rates (ANOVA and Scheffé tests) revealed that the 3 groups did not differ in the average number of baseline rotations when tested at 100–200 μA. However, the SWITCHERS made fewer baseline rotations than NONSWITCHERS at 250 μA ($P < 0.02$) and turned less than either controls or NONSWITCHERS at 300 μA ($P < 0.04$ and 0.009, respectively).

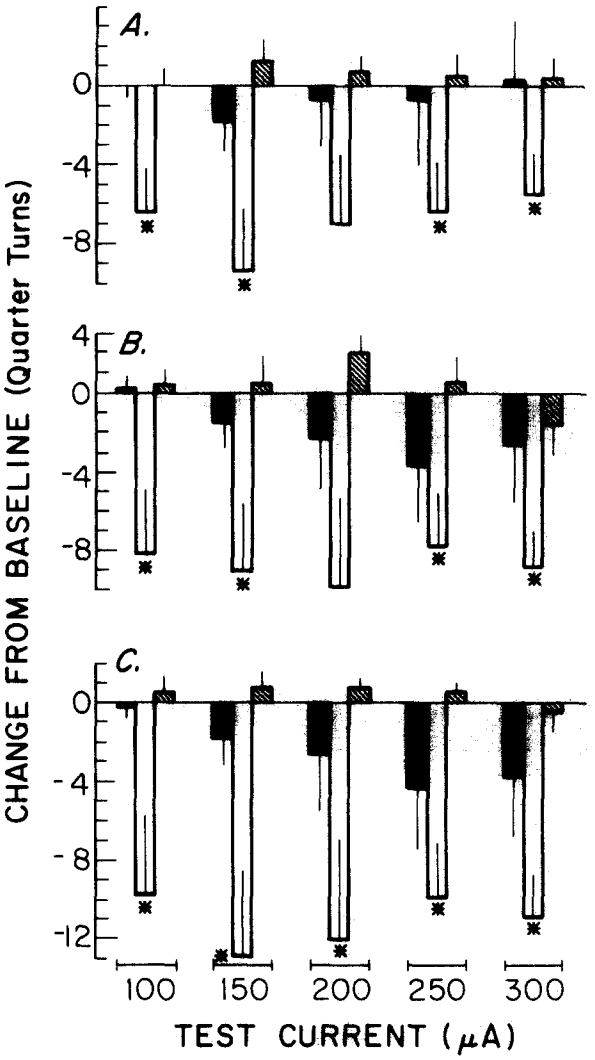

Fig. 2. Effects of 40 mg/kg of α-methyl-*p*-tyrosine on the contraversive ESRB of NONSWITCHERS and controls, and the ipsiversive ESRB of SWITCHERS tested with current intensities ranging from 100–300 μA (see text). Bars represent the average change (± the S.E.M.) in the number of one-quarter turns 2(A.), 4(B.), and 6(C.) hours after α-methyl-*p*-tyrosine treatment. Negative values indicate a reduction in rotational behavior. Open bars = NONSWITCHERS; solid bars = controls; hatched bars = SWITCHERS; *, differs from zero, $P < 0.05$ (two-tailed *t*-tests). Note: only the contraversive ESRB of nonswitchers was significantly attenuated by α-methyl-*p*-tyrosine.

Effects on ESRB of further DA depletion produced by α-methyl-p-tyrosine

Immediately following testing for ESRB on the 8th day after a 6-OHDA or saline infusion all rats received 40 mg/kg of α-MPT, and were again tested for ESRB 2, 4 and 6 h later. To determine if this low dose of α-MPT influenced ESRB the number of 90° turns made prior to α-MPT (first test on day 8) was subtracted from the number made 2, 4 or 6 h after α-MPT (separately for each current intensity). If α-MPT had no effect on ESRB this difference score should equal zero. Two-tailed *t*-tests were conducted to determine if the difference scores differed significantly from zero.

Fig. 2 shows the effect of α-MPT on ESRB in control rats, SWITCHERS and NONSWITCHERS. Treatment with α-MPT attenuated the contraversive ESRB in NONSWITCHERS at 4 of 5 current intensities within 2 h ($t > 2.6$, $P < 0.03$). By 6 h after α-MPT contraversive ESRB was significantly depressed in NONSWITCHERS at all current intensities tested ($t > 2.33$, $P < 0.05$). In striking contrast, this dose of α-MPT had no effect on contraversive ESRB in control animals, or on the ipsiversive ESRB of SWITCHERS.

Brain dopamine concentrations

Table I shows that the infusion of 6-OHDA into the right substantia nigra depleted DA in the right striatum, olfactory tubercle and nucleus accumbens. In all cases the extent of the DA depletion was greater in the striatum than in the accumbens or tubercle. A comparison of SWITCHERS and NONSWITCHERS revealed that the extent of the DA depletion was much greater in SWITCHERS than in NONSWITCHERS, averaging 64.6% in NONSWITCHERS and 98.4% in SWITCHERS. In fact, there was no overlap between the groups in the extent of the striatal DA depletion. The largest percent DA depletion in a NONSWITCHER was 94.1% (range, 35–94.1%), and the smallest percent striatal DA depletion in a SWITCHER was 96.5% (range, 96.5–99.0%). There was no overlap between SWITCHERS and NONSWITCHERS in the percent DA depletion in nucleus accumbens or olfactory tubercle either.

To estimate the extent of the DA depletion produced by 40 mg/kg of α-MPT the DA concentration of the striatum, olfactory tubercle and nucleus accumbens were determined in experimentally naive rats and control rats either: (1) 6 h after the administration of α-MPT; or (2) 6 h after administration of the vehicle. Some of these latter animals received α-MPT 5 days earlier, but they did not differ from experimentally naive rats and therefore the data were pooled. Table I shows that 40 mg/kg of α-MPT produced an average DA depletion of 53.5% in the striatum, 60.00% in the olfactory tubercle and 60.6% in the nucleus accumbens.

Histology

Clear histological data were not available for all animals because of tissue damage incurred during the preparation for neurochemical analyses. However, 13 sites which elicited contraversive turning were analyzed, and all were in or near the ascending nigrostriatal bundle in the LH. There was no obvious segregation of electrode sites in this small sample between the SWITCHERS, NONSWITCHERS and control animals.

TABLE I

Average (± S.E.M.) dopamine concentrations (ng/mg wet tissue) in selected brain regions on the side ipsilateral (IPSI) and contralateral (CONTRA) to a 6-OHDA lesion of the substantia nigra

Values are also given for control animals (average of left plus right) 6 h after an i.p. injection of α-methyl-*p*-tyrosine (MPT; 40 mg/kg) or the vehicle. See text for a definition of SWITCHERS vs NONSWITCHERS.

Structure	*NONSWITCHERS*		*Percent depletion*	*SWITCHERS*		*Percent depletion*	*Controls*		*Percent depletion*
	CONTRA	*IPSI*		*CONTRA*	*IPSI*		*Vehicle*	*MPT*	
Striatum	9.73±0.44	3.44±0.80	64.6%	12.27±1.91	0.20±0.07	98.4%	9.90±0.70	4.61±0.36	53.5%
Olfactory tubercle	5.62±0.66	4.42±0.62	21.4%	5.80±0.74	1.59±0.10	72.6%	5.47±0.40	2.19±0.24	60.0%
Nucleus accumbens	9.00±0.60	7.06±0.38	21.6%	10.95±0.35	1.43±0.64	86.9%	9.27±0.53	3.64±0.30	60.6%

DISCUSSION

The purpose of this study was to determine if nigrostriatal DA neurons are necessary for the contraversive rotation elicited by electrical stimulation in the LH. The results are clear. Nearly total depletion (> 96%) of striatal DA by 6-OHDA infusion into the substantia nigra completely abolished contraversive ESRB. However, there was not a graded reduction in rotational behavior dependent on the extent of the DA depletion, but rather what appeared to be an all-or-none effect. Partial striatal DA depletion (< 96%) failed to attenuate contraversive ESRB at all. In fact, there was a tendency (non-significant) for enhanced ESRB following partial DA depletion, as noted previously[32]. This may be due to the well known compensatory reaction of the nigrostriatal system to partial damage[35]. Further support for the involvement of nigrostriatal DA neurons in contraversive ESRB is provided by the observation that a dose of α-MPT that failed to disrupt ESRB in control animals, significantly attenuated ESRB in rats already partially depleted of striatal DA by a previous 6-OHDA infusion (c.f. ref. 43).

With the addition of the data provided here there is now considerable evidence that electrical stimulation of DA fibers ascending through the LH causes rotational behavior by releasing DA in the striatum. This evidence includes the following facts. (1) There is an anatomically circumscribed region in the LH where electrical stimulation produces vigorous contraversive ESRB. Only sites in or near ascending DA fibers produce contraversive ESRB, and the most effective sites (i.e., those that produce the most turns with the lowest current intensity) are the closest to the nigrostriatal bundle[30]. Using a HRP tracing technique in combination with electrical stimulation Bandler et al.[6] (ref. 6 p. 22) reported that, 'contraversive circling is elicited only when the stimulating electrode is within the substantia nigra zona compacta pathway'. It should be noted that the area in the LH where electrical stimulation produces contraversive ESRB is much more circumscribed than where stimulation elicits general hyperactivity, self-stimulation or consummatory behaviors[9,19]. (2) There are many studies showing that in fact DA is released in the striatum by stimulation at sites capable of producing contraversive ESRB[3,28,40]. (3) A lesion through an electrode that is effective in producing contraversive ESRB depletes ipsilateral striatal DA and permits vigorous amphetamine-induced turning[2]. (4) Contraversive ESRB is attenuated by systemically administered DA antagonists[5,31,39]. Finally, (5) relatively specific destruction of nigrostriatal DA neurons with intracerebral 6-OHDA abolishes contraversive ESRB (present study).

The observation that nearly total DA depletion was required to produce behavioral effects is not unique to rotational behavior. There have been many reports that very extensive DA depletion (> 95%) is required to produce aphagia and adipsia, and that partial DA depletion has only transient and mild effects on regulatory behaviors (for review see ref. 35). It appears that if only 4–5% of the DA innervation in the striatum remains, completely normal levels of function are possible — as indicated by the vigor of rotational behavior elicited by electrical stimulation. This remarkable ability of the nigrostriatal system to adapt to extensive damage is usually attributed to its ability to generate both presynaptic[1,22] and postsynaptic[10,24,37,44] compensatory responses. However, it is difficult to believe that only 4–5% of the remaining fibers generate a compensatory response of such magnitude that they can perform the task of an intact nigrostriatal DA system; unless, only a small fraction of the fibers are usually required for 'normal' function. Perhaps, only a small percentage of nigrostriatal DA cells need be active at any point in time for full function. If so, this may help explain why considerable function is restored following grafts of embryonic or adrenal tissue that only innervate a small portion of the normal terminal field[12,17,18].

Following nearly total DA depletion animals not only ceased to show contraversive ESRB, but within 2–3 days showed stable and vigorous ipsiversive ESRB. Christie et al.[8] previously reported a somewhat similar finding in a short abstract. Obviously, this ipsiversive ESRB was not dependent on activation of nigrostriatal DA neurons because: (1) it occurred in those rats with the most extensive DA depletion; and (2) ipsiversive ESRB was not influenced by subsequent treatment with α-MPT. In a previous study[30] we observed ipsiversive ESRB in non-lesioned rats following stimulation near the crus cerebri, close to the nigrostriatal bundle in the far lateral hypothala-

mus. Others have reported that stimulation of similar fibers further posterior, in the cerebral peduncle, also produces ipsiversive rotational behavior that is not attenuated by DA antagonists[39]. Therefore, the most likely explanation of why some animals switched from contraversive to ipsiversive turning is because the effect of current spread to the crus cerebri was 'unmasked' following total destruction of ascending DA fibers. The origin and destination of the fibers in the crus cerebri responsible for ipsiversive ESRB is not known. It is possible striatonigral fibers are involved because stimulation of the lateral substantia nigra also produces ipsiversive ESRB[38].

Why contraversive ESRB elicited by stimulation in LH is attenuated by striatal DA depletion, but contraversive ESRB elicited by stimulation in the substantia nigra is not[32] remains an open question. The most obvious possibility is that the intrahypothalamic 6-OHDA injection used by Saranak and Goldfarb[32] did not sufficiently deplete striatal DA. Saranak and Goldfarb[32] reported an average depletion of striatal DA of 93%, and we found that a 96% depletion is necessary to abolish contraversive ESRB. However, arguing against this possibility is the fact that Saranak and Goldfarb[32] reported that in 3 of their rats striatal DA was undetectable. Since these authors used a sensitive gas chromatographic technique to assay DA, these latter 3 rats probably had a nearly complete striatal DA depletion — yet presumably they continued to show contraversive ESRB (only averaged data are presented).

Another possibility is that the contraversive ESRB elicited by stimulation of the substantia nigra has a different neural basis than that elicited by stimulation in the LH. It has been suggested that nigral stimulation may produce contraversive rotational behavior by activation of neurons postsynaptic to nigrostriatal DA cells, perhaps striatonigral projections[32,33]. If this is the case, the ipsiversive ESRB seen with stimulation of the crus cerebri is probably not due to activation of the same striatonigral fibers. Stimulation of the nigra also activates a direct descending pathway that presumably relays in the reticular formation and is responsible for the enhancement of spinal monosynaptic reflexes[41]. Therefore, the possibility that some of the behavioral effects elicited by nigral stimulation are due to activation of nigral-reticular-spinal motor circuits should be considered. This idea is supported by the observation that rats show turning behavior in the absence of the entire telencephalon[26,27]. Furthermore, the enhancement of lumbo-sacral monosynaptic reflexes is antagonized by haloperidol and chlorpromazine, suggesting a DA link in the reticular formation[42]. This might explain why nigral-induced rotation is not attenuated by intrahypothalamic 6-OHDA[32], but is attenuated by the systemic administration of DA antagonists[31,39].

Given the data presently available one could also argue that ascending fibers are involved in mediating the rotational behavior evoked by both hypothalamic and nigral stimulation, because electrolytic lesions of the LH, caudate or globus pallidus do attenuate nigral-evoked contraversive ESRB[32,33]. It is well established that the striatum is anatomically and functionally heterogeneous[11,13–15]. Perhaps there is a specific striatal subregion that must be denervated to abolish contraversive ESRB, and this region was not sufficiently damaged by intrahypothalamic 6-OHDA[32,33]; but was by intranigral 6-OHDA (present study). Of course, one would have to assume that electrolytic lesions of the LH did damage the fibers innervating this critical subregion[32]. Reports that grafts of embryonic nigral tissue that innervate the dorsal striatum produce recovery from rotational asymmetries, whereas grafts that innervate the lateral and ventral striatum do not[12] support this possibility (but also see ref. 15).

In summary, this experiment supports the hypothesis that the contraversive rotational behavior evoked by LH stimulation is due to activation of dopaminergic cells innervating the forebrain[2,4,5]. Whether the contraversive ESRB evoked by hypothalamic stimulation and that elicited by nigral stimulation have a similar or different neural basis remains to be determined. We propose that the contraversive ESRB evoked by electrical stimulation of the nigrostriatal bundle in the LH provides a more appropriate model for studying the functional activity of the nigrostriatal system (e.g. ref. 30), than does rotational behavior elicited by nigral stimulation; in part because of the complexity of interpreting the effects of stimulation in this latter region (e.g. refs. 38 and 39).

ACKNOWLEDGEMENTS

This research was supported by Grant 16437 from

the NINCDS. Dr. Becker was supported by Grant 05997 from the NICHHD, and E. Castañeda by a University of Michigan Rackham Minority Fellowship. We thank Marylin Hoy for secretarial services and Merrell-Dow Pharmaceuticals for the gift of the desipramine.

REFERENCES

1 Agid, Y., Javoy, F. and Glowinski, J., Hyperactivity of remaining dopaminergic neurones after partial destruction of the nigro-striatal dopaminergic system in the rat, *Nature New Biol.*, 245 (1973) 150–151.
2 Arbuthnott, G. W. and Crow, T. J., Relation of contraversive turning to unilateral release of dopamine from the nigrostriatal pathway in rats, *Exp. Neurol.*, 30 (1971) 484–491.
3 Arbuthnott, G. W., Crow, T. J., Fuxe, K., Olson, L. and Ungerstedt, U., Depletion of catecholamines in vivo induced by electrical stimulation of central monoamine pathways, *Brain Research*, 24 (1970) 471–483.
4 Arbuthnott, G. W. and Ungerstedt, U., Locomotor behaviour after electrical stimulation of dopamine containing neurones, *Acta physiol. scand.*, 77, Suppl. 330 (1969) 117.
5 Arbuthnott, G. W. and Ungerstedt, U., Turning behavior induced by electrical stimulation of the nigro-neostriatal system of the rat, *Exp. Neurol.*, 47 (1975) 162–172.
6 Bandler, R., Tork, I. and Cher, L., Anatomical demonstration of the involvement of the substantia nigra in contraversive circling elicited by electrical stimulation of the medial forebrain bundle in the cat: a retrograde transport study using horseradish peroxidase subsequent to an electrolytic lesion, *Neurosci. Lett.*, 26 (1981) 17–23.
7 Breese, G. R. and Traylor, T. D., Depletion of brain noradrenaline and dopamine by 6-hydroxydopamine, *Brit. J. Pharmacol.*, 42 (1971) 88–89.
8 Christie, J. E., Ljungberg, T. and Ungerstedt, U., Dopamine neurones and electrical self-stimulation in the lateral hypothalamus, *J. Physiol. (Lond.)*, 234 (1973) 80P–81P.
9 Cox, V. C. and Valenstein, E. S., Distribution of hypothalamic sites yielding stimulus-bound behavior, *Brain Behav. Evol.*, 2 (1969) 359–376.
10 Creese, I., Burt, D. R. and Snyder, S. H., Dopamine receptor binding enhancement accompanies lesion-induced behavioral supersensitivity, *Science*, 197 (1977) 596–598.
11 Divac, I., Rosvold, H. E. and Szwarcbart, M. K., Behavioral effects of selective ablation of the caudate nucleus, *J. comp. Physiol. Psychol.*, 63 (1967) 184–190.
12 Dunnett, S. B., Bjorklund, A., Stenevi, U. and Iversen, S. D., CNS transplantation: structural and functional recovery from brain damage. In R. M. Buijs, P. Pevet and D. F. Swaab (Eds.), *Chemical Transmission in the Brain, Progress in Brain Research, Vol. 55*, Elsevier, Amsterdam, 1982, pp. 431–443.
13 Dunnett, S. B. and Iversen, S. D., Regulatory impairments following selective 6-OHDA lesions of the neostriatum, *Behav. Brain Res.*, 4 (1982) 195–202.
14 Dunnett, S. B. and Iversen,S. D., Sensorimotor impairments following localized 6-hydroxydopamine and kainic acid-induced lesions of the neostriatum, *Brain Research*, 248 (1982) 121–127.
15 Dunnett, S. B. and Iversen, S. D., Spontaneous and drug-induced rotation following localized 6-hydroxydopamine and kainic acid-induced lesions of the neostriatum, *Neuropharmacology*, 21 (1982) 899–908.
16 Felice, L. J., Felice, J. D. and Kissinger, P. T., Determination of catecholamines in rat brain parts by reverse-phase ion-pair chromatography, *J. Neurochem.*, 31 (1978) 1461–1465.
17 Freed, W. J., Perlow, M. J., Karoum, F., Seiger, A., Olson, L., Hoffer, B. J. and Wyatt, R. J., Restoration of dopaminergic function by grafting of fetal rat substantia nigra to the caudate nucleus: long-term behavioral, biochemical, and histochemical studies, *Ann. Neurol.*, 8 (1980) 510–519.
18 Freed, W. J., Morihisa, J. M., Spoor, E., Hoffer, B. J., Olson, L., Seiger, A. and Wyatt, R. J., Transplanted adrenal chromaffin cells in rat brain reduce lesion-induced rotational behaviour, *Nature (Lond.)*, 292 (1981) 351–352.
19 German, D. C. and Bowden, D. M., Catecholamine systems as the neural substrate for intracranial self-stimulation: a hypothesis, *Brain Research*, 73 (1974) 381–419.
20 Glick, S. D., Jerussi, T. P. and Fleisher, L. N., Turning in circles: the neuropharmacology of rotaton, *Life Sci.*, 18 (1976) 889–896.
21 Glick, S. D., Jerussi, T. P. and Zimmerberg, B., Behavioral and neuropharmacological correlates of nigrostriatal asymmetry in rats. In S. Harnad, R. W. Doty, L. Goldstein, J. Jaynes and G. Krauthamer (Eds.), *Lateralization in the Nervous System*, Academic Press, New York, 1977, pp. 213–249.
22 Hefti, F., Melamed, E. and Wurtman, R. J., Partial lesions of the dopaminergic nigrostriatal system in rat brain: biochemical characterization, *Brain Research*, 195 (1980) 123–137.
23 Jerussi, T. P. and Glick, S. D., Amphetamine-induced rotation in rats without lesions, *Neuropharmacology*, 13 (1974) 283–286.
24 Mishra, R. K., Gardner, E. L., Katzman, R. and Makman, M. H., Enhancement of dopamine-stimulated adenylate cyclase activity in rat caudate after lesions in substantia nigra: evidence for denervation supersensitivity, *Proc. nat. Acad. Sci. (U.S.A.)*, 71 (1974) 3883–3887.
25 Morrison, D. F., *Multivariate Statistical Methods*, McGraw-Hill, New York, 1967.
26 Papadopoulos, G. and Huston, J. P., Contralateral turning after unilateral electrolytic lesion of substantia nigra in thalamic rats, *Neurosci. Lett.*, 13 (1979) 63–67.
27 Papadopoulos, G. and Huston, J. P., Removal of the telencephalon spares turning induced by injection of GABA agonists and antagonists into the substantia nigra, *Behav. Brain Res.*, 1 (1980) 25–38.
28 Portig, P. J. and Vogt, M., Release into the cerebral ventricles of substances with possible transmitter functions in the caudate nucleus, *J. Physiol. (Lond.)*, 204 (1969) 687–715.
29 Pycock, C. J., Turning behaviour in animals, *Neuroscience*, 5 (1980) 461–514.
30 Robinson, T. E., Camp, D. M., Jacknow, D. S. and Becker, J. B., Sex differences and estrous cycle dependent variation in rotational behavior elicited by electrical stimulation

of the mesostriatal dopamine system, *Behav. Brain Res.*, 6 (1982) 273–287.

31 Roffman, M., Bernard, P. S., Dawson, K. M., Sobiski, R. E. and Saelens, J. K., The effects of haloperidol and clozapine on circling induced by electrical stimulation of the substantia nigra and the ventromedial tegmentum, *Neuropharmacology*, 17 (1978) 943–946.

32 Saranak, J. and Goldfarb, J., Effects of electrolytic and 6-hydroxydopamine lesions of the lateral hypothalamus on rotation evoked by electrical stimulation of the substantia nigra in rats, *Brain Research*, 208 (1981) 81–95.

33 Saranak, J. and Goldfarb, J., Rotation evoked by nigral stimulation following lateral hypothalamic, striatal, and pallidal lesions in rats, *Exp. Neurol.*, 77 (1982) 295–313.

34 Spector, S., Sjoerdsma, A. and Udenfriend, S., Blockade of endogenous norepinephrine synthesis by α-methyl-tyrosine, an inhibitor of tyrosine hydroxylase, *J. Pharmacol. exp. Ther.*, 147 (1965) 86–95.

35 Stricker, E. M. and Zigmond, M. J., Recovery of function after damage to central catecholamine-containing neurons: a neurochemical model for the lateral hypothalamic syndrome. In J. M. Sprague and A. Epstein (Eds.), *Progress in Psychobiology and Physiological Psychology, Vol. 6*, Academic Press, New York, 1976, pp. 121–187.

36 Ungerstedt, U., Striatal dopamine release after amphetamine or nerve degeneration revealed by rotational behavior, *Acta physiol. scand.*, 82, Suppl. 367 (1971) 49–68.

37 Ungerstedt, U., Postsynaptic supersensitivity after 6-hydroxydopamine induced degeneration of the nigro-neostriatal dopamine system in the rat brain, *Acta physiol. scand.*, 82, Suppl. 367 (1971) 69–93.

38 Vaccarino, F. J. and Franklin, K. B. J., Self-stimulation and circling reveal functional differences between medial and lateral substantia nigra, *Behav. Brain Res.*, 5 (1982) 281–295.

39 Vaccarino, F. J. and Franklin, K. B. J., Dopamine mediates ipsi- and contraversive circling elicited from the substantia nigra, *Pharmacol. Biochem. Behav.*, 17 (1982) 431–434.

40 Von Voigtlander, P. F. and Moore, K. E., The release of ^{3}H-dopamine from cat brain following electrical stimulation of the substantia nigra and caudate nucleus, *Neuropharmacology*, 10 (1971) 733–741.

41 York, D. H., Potentiation of lumbo-sacral monosynaptic reflexes by the substantia nigra, *Exp. Neurol.*, 36 (1972) 437–448.

42 York, D. H., Antagonism of descending effects of the substantia nigra on lumbo-sacral monosynaptic reflexes, *Neuropharmacology*, 12 (1973) 629–636.

43 Zigmond, M. J. and Stricker, E. M., Deficits in feeding behavior after intraventricular injection of 6-hydroxydopamine in rats, *Science*, 177 (1972) 1211–1214.

44 Zigmond, M. J. and Stricker, E. M., Compensatory changes after intraventricular administration of 6-hydroxydopamine: a neurochemical model for recovery of function. In T. Malmfors, O. Almgren, A. Carlsson, J. Engel, G. Jonsson and C. Sachs (Eds.), *Chemical Tools in Catecholamine Research*, North Holland, Amsterdam, 1975.