

## A QUANTITATIVE ROTATIONAL MODEL FOR STUDYING SEROTONERGIC FUNCTION IN THE RAT

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(Accepted July 19th, 1976)

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### SUMMARY

Unilateral injection of 5,7-dihydroxytryptamine (4  $\mu\text{g}/4 \mu\text{l}$ ) into the medial forebrain bundle of rats produced serotonin depletions of 65% and 70% in the ipsilateral corpus striatum and ipsilateral forebrain, respectively. These animals showed a dose-dependent increase in contralateral turning (rotational behavior) when pre-treated with a peripheral decarboxylase inhibitor and then injected with L-5-hydroxytryptophan in doses ranging from 5 to 100 mg/kg i.p. Injections of *p*-chloroamphetamine, which releases endogenous stores of serotonin, produced ipsilateral turning which could be blocked by prior serotonin depletion. Systemic administration of the catecholamine drugs L-DOPA, apomorphine and D-amphetamine never elicited consistent turning in either direction in these animals. These data indicate that the turning response of rats with unilateral destruction of brain serotonin nerve terminals provides a sensitive tool for quantifiably studying changes in serotonergic function.

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### INTRODUCTION

Although pharmacological and neurochemical studies have provided a wealth of information about CNS neurotransmitter systems, it is behavior that represents the ultimate probe into the functional activity of these systems. The utility of behavioral indices of synaptic function is perhaps best exemplified by the impressive advances in our understanding of brain dopamine that have accrued from the employment of one particular model. When portions of the dopaminergic nigrostriatal system are destroyed or damaged unilaterally, compounds which either increase synaptic dopamine or directly stimulate postsynaptic dopamine receptors produce unilateral turning<sup>1,23,25,27</sup>. The directionality of the turning is dependent on whether the test compound has a direct postsynaptic effect or whether it acts presynaptically. Compounds with a direct postsynaptic effect, such as apomorphine<sup>2,8</sup>, produce contralateral turning

which is hypothesized to be attributable to its increased efficacy on the denervated striatal neurons, i.e., denervation supersensitivity<sup>24,27</sup>. On the other hand, compounds which release presynaptic stores of dopamine, such as amphetamine<sup>5,11</sup>, produce ipsilateral turning which is hypothesized to be due to the increased release of dopamine stores on the intact side as compared to the denervated side<sup>23,27</sup>. The present report describes the development of an analogous turning or rotational model for studying the activity of synaptic serotonin in the CNS.

Rotational models have two major advantages. First, in addition to providing a qualitative index of change in a particular transmitter system, they provide a quantitative estimate of synaptic function since the number of turns per unit time increases as a function of dose of the test compound. Second, they provide an extremely sensitive measure of activity in the system. For example, injections of apomorphine can elicit contralateral turning in doses far below those required to produce either increased locomotor activity or response stereotypy in normal animals<sup>23,24,27</sup>. Similarly, we have previously described an animal behavior model for studying synaptic serotonin, but one that is sensitive to only large increases in CNS serotonin, e.g., that produced by 150–200 mg/kg L-5-hydroxytryptophan (L-5-HTP)<sup>13</sup>, whereas the present rotational model reflects change in CNS serotonin produced by doses of L-5-HTP as low as 5 or 10 mg/kg.

The present studies examined the influence of both serotonergic and catecholaminergic drugs on the induction of turning in rats with unilateral destruction of either central serotonergic or catecholaminergic nerve terminals.

#### METHODS

Male Sprague–Dawley rats weighing 290–330 g at the time of surgery were used in all experiments. Neurochemical destruction of serotonin or catecholamine nerve terminals was accomplished by intracerebral injections of 5,7-dihydroxytryptamine (5,7-DHT) or 6-hydroxydopamine (6-OHDA), respectively. In all cases the compounds were injected unilaterally, on the left side of the brain. Forty-five minutes prior to the injection of 5,7-DHT creatinine sulfate (4  $\mu$ g of base in 4  $\mu$ l of sterile saline with 0.2 mg/ml ascorbic acid added) animals were pretreated with desipramine hydrochloride (25 mg/kg i.p. dose as salt) in order to prevent destruction of catecholamine nerve terminals<sup>4,10</sup>. The animals were anesthetized with sodium pentobarbital (50 mg/kg i.p.) and placed in a stereotaxic instrument with the bite bar oriented 5 mm above the interaural line. Injections of the 4  $\mu$ l solution were made over a 15 min period via a 32-gauge cannula whose tip was localized at AP + 4.8, ML + 1.7, and DV — 2.1. The cannula was left in place for 4 min after the injection to permit diffusion of the 5,7-DHT. Injections of 6-OHDA hydrobromide (6  $\mu$ g of base in 4  $\mu$ l of sterile saline with 0.2 mg/ml ascorbic acid added) were made under ether anesthesia. The animals were placed in a stereotaxic instrument with the bite bar oriented 4 mm below the interaural line. Injections were carried out as described for 5,7-DHT but the target coordinates were AP + 7.3, ML + 2.5, and DV — 6.9 from dura. After removal of the injection cannula, the scalp was sutured and animals were administered 30,000 units of Duracillin i.m.

Behavioral testing was not begun until at least two weeks postsurgery. The testing procedure was similar to that previously described by Ungerstedt and Arbuthnott<sup>25</sup>. Animals were run in hemispheric plastic bowls (36 cm in diameter and 18 cm deep) painted flat black and containing a layer of sawdust. A broad rubber band attached to an overhead swivel by means of a stiff wire was placed around the mid-section of the animal. Each time the animal made a full 360° turn around the bowl a microswitch was closed either to the left or to the right, depending on the direction of movement. The number of left and right closures were automatically counted. Following a brief habituation period, the number of turns during a 15 min baseline were recorded. Animals were then administered the experimental drug and the number of turns occurring during the next hour were tallied every 15 min. All data analyses were performed on the number of net turns (ipsilateral minus contralateral, or vice versa) occurring between 15 and 60 min postinjection. Experiments were run in a well-ventilated room with overhead fluorescent lights and a 70 dB masking noise.

The following drugs were administered via intraperitoneal injection to both 5,7-DHT and 6-OHDA lesioned animals: L-5-hydroxytryptophan (5, 10, 25, 50 and 100 mg/kg), L-DOPA (5, 10, 25 and 50 mg/kg — the 100 mg/kg dose proved to be too toxic), apomorphine hydrochloride (0.1 and 0.2 mg/kg as salt), D-amphetamine sulfate (0.5 and 1.0 mg/kg as salt) and D,L-*p*-chloroamphetamine hydrochloride (PCA) (2.5 mg/kg as salt). PCA was also tested in 5,7-DHT and 6-OHDA lesioned animals pretreated with D,L-*p*-chlorophenylalanine methyl ester hydrochloride (PCPA) (300 mg/kg as salt 3 days before). One hour prior to administration of either L-5-hydroxytryptophan or L-DOPA, animals were given an injection of a peripherally acting L-amino acid decarboxylase inhibitor (MK 486, 75 mg/kg i.p.) in order to minimize any peripheral side effects of the metabolites of these compounds. All drugs were dissolved in saline. MK 486, L-5HTP and L-DOPA also required small amounts of HCl, which was also added to the saline control injections.

At the completion of the study, subgroups of the 5,7-DHT and 6-OHDA treated rats were selected for monoamine assays. In addition, the brains of a group of 5,7-DHT treated animals that did not show consistent contralateral turning in response to 5-HTP were also assayed. Serotonin, norepinephrine and dopamine were measured using a method developed by Jacobowitz and his colleagues<sup>18</sup>. Rats were decapitated and the brains were removed and dissected on ice, obtaining the left and right corpora striata and the rest of the forebrain (obtained by making a coronal cut immediately caudal to mammillary bodies and one in the midsagittal plane). The tissues were immediately frozen in liquid nitrogen, and were assayed within 8 h. The tissues were weighed and placed in 5 ml of ice-cold n-butanol. Assuming tissue to be 70% water, 0.01 N HCl was added to make 0.75 ml total water in the 5 ml of butanol. The tissue was homogenized thoroughly and the homogenate transferred to plastic centrifuge tubes. For serotonin assays, two ml of the 3000 × g supernatant were added to stoppered centrifuge tubes containing 5 ml of n-heptane and 0.5 ml of 0.1 N HCl. After vortexing for 20 sec and centrifuging at 3000 rev./min for 5 min, the organic phase was removed by aspiration. Aliquots of the acid phase (0.3 ml) were transferred to test tubes and 0.2 ml of OPT solution (50 mg of ortho-phthalaldehyde in 100 mg of

absolute methanol) and 1.5 ml of 10 *N* HCl were added. After heating in boiling water for 10 min and cooling to room temperature, fluorescence was read at 470 nm with activation at 360 nm. The samples were read against external standards of serotonin (0.03–0.3 nmole) and reagent blanks.

For catecholamine assays, 2 ml of the butanol supernatant were added to stoppered centrifuge tubes containing 2 ml of 0.1 *M* sodium phosphate buffer (pH 6.5). After vortexing for 20 sec and centrifuging at 3000 rev./min for 10 min, the organic phase was removed by aspiration. Aliquots of the buffer (1.0 ml) were transferred to test tubes containing 0.25 ml of versene (4.0 g EDTA in 100 ml distilled water with pH adjusted to 6.5 with 10 *N* NaOH). Then the following solutions were added at 3-min intervals: 0.20 ml iodine solution (4.8 g potassium iodide plus 0.25 g iodine in 100 ml distilled water); 0.25 ml alkaline sulfite solution (2.5 g sodium sulfite in 100 ml of 4 *N* NaOH); and 0.30 ml of 5 *N* acetic acid. The tubes were heated in boiling water for 5 min and cooled to room temperature. Norepinephrine fluorescence was read immediately at 485 nm with activation at 385 nm, and dopamine fluorescence was read 30 min later at 380 nm with activation at 322 nm<sup>6</sup>. The samples were read against external standards of norepinephrine and dopamine (0.02–0.2 nmole) as well as reagent and tissue blanks.

The monoamine values have not been corrected for recoveries, which were as follows: serotonin, 94.9 ± 2.1%; norepinephrine, 97.3 ± 2.8% and dopamine, 95.8 ± 3.1%.

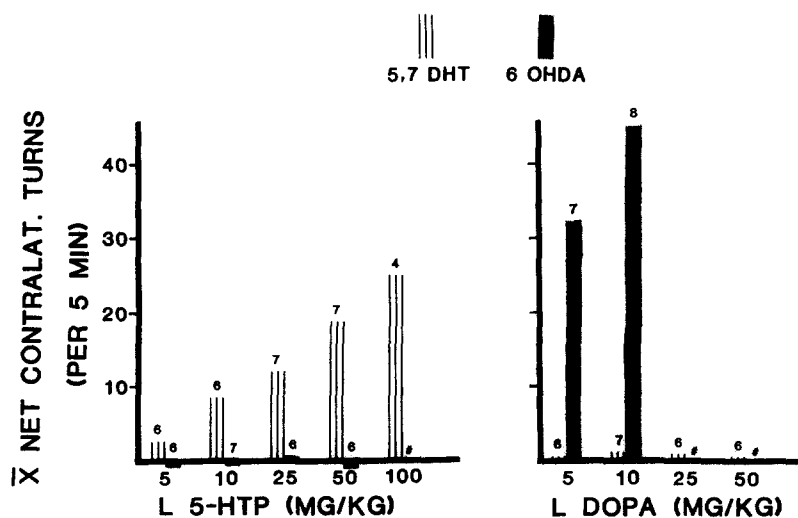


Fig. 1. Effect of various doses of L-5-HTP (left panel) and L-DOPA (right panel) on the elicitation of turning in 5,7-DHT and 6-OHDA lesioned rats. All animals were administered a peripheral decarboxylase inhibitor (MK 486, 75 mg/kg, i.p.) 1 h prior to L-5-HTP or L-DOPA. Bars extending above the abscissa represent mean net contralateral turns, while those extending below the baseline represent mean net ipsilateral turns. Numbers above each bar represent the subjects per group, and # indicates that the particular group was not run (see text for explanation). Note the minimal response to L-DOPA in 5,7-DHT lesioned animals, and the minimal response to 5-HTP in 6-OHDA lesioned animals.

## RESULTS

Unilateral 5,7-DHT or 6-OHDA lesions did not produce any marked tendency for untreated animals to turn unilaterally. When 5,7-DHT animals were injected with saline, they demonstrated no particular propensity to turn either ipsilateral or contralateral to the lesion. However, when 6-OHDA lesioned animals were injected with saline, they showed a slight, but consistent, tendency to turn to the side ipsilateral to the lesion ( $\bar{X} \cong 1-2$  net ipsilateral turns/5 min).

Administration of L-5-HTP to 5,7-DHT lesioned rats pretreated with a peripheral decarboxylase inhibitor, MK-486 (75 mg/kg i.p.), resulted in a significant dose-dependent increase in net contralateral turns ( $F = 22.6$ ; 4,40 *df*;  $P < 0.001$ ) (Fig. 1, left panel). This effect ranged from a mean of 2.4 turns/5 min at a dose of 5 mg/kg i.p. to 25.4 turns/5 min at a dose of 100 mg/kg i.p. By contrast, administration of L-DiOPA to these same animals (also pretreated with MK-486) never produced consistently unilateral turning, even in doses as high as 50 mg/kg i.p. ( $P > 0.10$ ) (Fig. 1, right panel). The 100 mg/kg dose of L-DOPA was discontinued because it greatly debilitated the animals and in some cases proved fatal.

Examination of the data for 6-OHDA lesioned animals shows a response pattern reciprocal to that seen in 5,7-DHT lesioned animals. They showed no turning in response to L-5-HTP and a dramatic increase in turning in response to L-DOPA. No consistent pattern of turning was seen in 6-OHDA lesioned rats pretreated with MK-486 and given L-5-HTP in doses up to 50 mg/kg i.p. ( $P > 0.10$ ) (Fig. 1, left panel). The 100 mg/kg dose of 5-HTP was not administered because it was clear that in general 5-HTP would not produce a dose-related turning effect, and because these high drug doses were often toxic. When L-DOPA was administered to 6-OHDA lesioned animals pretreated with MK-486, it produced significant contralateral turning in doses as low as 5 and 10 mg/kg i.p. ( $\bar{X} = 33.3$  and 45.6 net contralateral turns/5 min, respectively;  $F = 143.5$ ; 1,10 *df*;  $P < 0.001$ ) (Fig. 1, right panel). Higher doses of L-DOPA were not administered to these animals because of the clear-cut effects at the low doses, and because of the toxic effects of high doses of L-DOPA.

It is well known that the dopamine precursor, L-DOPA, elicits contralateral turning in rats with unilateral 6-OHDA lesions, and that the catecholamine releaser, D-amphetamine, elicits ipsilateral turning in these animals. In an attempt to determine whether this relationship would be paralleled in 5,7-DHT lesioned animals, we examined the effects of the serotonin releaser, PCA<sup>22,29</sup>, to see if it would elicit ipsilateral turning (Table I). In a dose of 2.5 mg/kg i.p., PCA elicited strong and consistent ipsilateral turning ( $\bar{X} = 9.4$  net ipsilateral turns/5 min). This effect was completely blocked by prior serotonin synthesis inhibition with PCPA ( $\bar{X} = 0.3$  net ipsilateral turns/5 min). When the same dose of PCA was administered to 6-OHDA lesioned animals, it also elicited strong ipsilateral turning ( $\bar{X} = 26.1$  net turns/5 min), but this effect, although diminished, was not blocked by prior serotonin synthesis inhibition ( $\bar{X} = 16.0$  net ipsilateral turns/5 min).

In an attempt to further examine the specificity of the turning induced in 5,7-DHT lesioned animals, they were administered the catecholamine releaser, D-amphet-

TABLE I

*Effects of various drugs on inducing turning in unilateral 5,7-DHT and 6-OHDA lesioned rats*

Drug	Dose (mg/kg)	N	$\bar{X}$ turns/5 min $\pm$ S.E.M.***		
			5,7-DHT		6-OHDA
Saline	—	7	—0.5 $\pm$ 0.8	7	+1.4 $\pm$ 0.4
PCA	2.5	7	+9.4 $\pm$ 1.7**	4	+26.1 $\pm$ 4.2*
PCA + PCPA	2.5	8	+0.3 $\pm$ 0.4	5	+16.0 $\pm$ 2.5*
Apomorphine	0.1	8	0.0 $\pm$ 0.4	5	—46.1 $\pm$ 6.6**
Apomorphine	0.2	8	+0.2 $\pm$ 0.1	6	—35.3 $\pm$ 7.7**
Amphetamine	0.5	8	+1.0 $\pm$ 0.4	7	+11.2 $\pm$ 3.7*
Amphetamine	1.0	8	—2.4 $\pm$ 1.0	5	+33.3 $\pm$ 7.3*

\* Significantly different from saline control,  $P < 0.05$  (Dunnett's test).

\*\*  $P < 0.001$  (Dunnett's Test).

\*\*\* +, net ipsilateral turns; —, net contralateral turns.

amine (0.5 and 1.0 mg/kg i.p.) and the dopamine agonist, apomorphine (0.1 and 0.2 mg/kg i.p.). Neither of these drugs produced any strong or consistent turning in 5,7-DHT lesioned animals (Table I). On the other hand, when they were administered to 6-OHDA lesioned animals, D-amphetamine elicited strong ipsilateral turning (e.g.  $\bar{X} = 33.3$  net turns/5 min with 1.0 mg/kg i.p.), and apomorphine elicited strong contralateral turning (e.g.  $\bar{X} = 46.1$  net turns/5 min with 0.1 mg/kg i.p.).

The ratio of the number of turns to the preferred side over the number of turns to the non-preferred side was typically between 8:1 and 12:1, both across the two lesion groups and across the various drug treatments. The single exception to this was an approximately 30:1 turning ratio for the effects of apomorphine in 6-OHDA lesioned animals.

Neurochemical analyses displayed in Table II indicate that injection of 5,7-DHT into the left medial forebrain bundle significantly decreased the serotonin content of the left corpus striatum to 34.8% of that in the right corpus striatum. Similarly, the serotonin content of the left side of the forebrain was decreased to 30.6% of that in the right side. There was a smaller, but statistically significant decrease of norepinephrine in the left striatum, but not in the left forebrain. No statistically significant change in either left striatal or left forebrain dopamine was produced by 5,7-DHT injections. Analyses of the brains of 5,7-DHT lesioned animals that did not display consistent turning in response to L-5-HTP showed that the reduction in serotonin content of the left striatum, although statistically significant, was only 38.1% as opposed to the 65.2% decrease found in the consistent turners. Similarly, the decrease in serotonin content of the left forebrain was 56.8% in the inconsistent turners as opposed to 69.4% in the consistent turners. No significant decreases were observed in dopamine or norepinephrine levels in either the left striatum or left forebrain of the inconsistent turners.

Injection of 6-OHDA into the nigrostriatal pathway produced marked reductions in dopamine and norepinephrine content of both the left striatum and left

TABLE II  
Effect of unilateral (left side) brain injections of 5,7-DHT or 6-OHDA on striatal and forebrain monoamine levels ( $\bar{X} \pm S.E.M.$ ), in  $\mu\text{g/g}$  tissue

Group	N	Corpus striatum			Forebrain		
		5-HT	NE	DA	5-HT	NE	DA
5,7-DHT	7						
Consistent turners							
Right		1.535 $\pm$ 0.136	0.320 $\pm$ 0.024	4.035 $\pm$ 0.176	0.538 $\pm$ 0.011	0.373 $\pm$ 0.025	0.397 $\pm$ 0.028
Left		0.535 $\pm$ 0.086*	0.243 $\pm$ 0.026*	3.993 $\pm$ 0.331	0.165 $\pm$ 0.018*	0.336 $\pm$ 0.032	0.336 $\pm$ 0.023
(%)		(34.8%)	(75.9%)	(98.9%)	(30.6%)	(90.0%)	(84.6%)
Inconsistent turners	4						
Right		1.467 $\pm$ 0.271	0.276 $\pm$ 0.033	4.153 $\pm$ 0.249	0.532 $\pm$ 0.065	0.377 $\pm$ 0.031	0.438 $\pm$ 0.052
Left		0.909 $\pm$ 0.201*	0.280 $\pm$ 0.071	3.908 $\pm$ 0.337	0.230 $\pm$ 0.017*	0.373 $\pm$ 0.047	0.379 $\pm$ 0.071
(%)		(61.9%)	(101.4%)	(94.1%)	(43.2%)	(98.9%)	(82.4%)
6-OHDA	4						
Right		1.534 $\pm$ 0.138	0.316 $\pm$ 0.009	4.461 $\pm$ 0.206	0.537 $\pm$ 0.026	0.341 $\pm$ 0.027	0.389 $\pm$ 0.009
Left		1.543 $\pm$ 0.117	0.045 $\pm$ 0.015*	0.130 $\pm$ 0.027*	0.539 $\pm$ 0.025	0.070 $\pm$ 0.016*	0.058 $\pm$ 0.009*
(%)		(100.6%)	(14.2%)	(2.9%)	(100.4%)	(20.5%)	(14.9%)

\*  $P < 0.001$  (two-tailed  $t$ -tests, comparing right and left sides). Significance level was set at  $P < 0.01$  because of the use of multiple  $t$ -tests.

forebrain (Table II). The treatment produced virtually total depletion of left striatal dopamine (2.9% of the content of the right striatum).

## DISCUSSION

Examination of turning behavior in rats following unilateral destruction specific to forebrain serotonin nerve terminals by injections of 5,7-DHT into the medial forebrain bundle provides a viable method for studying serotonergic function. Consistent, dose-dependent turning to the side contralateral to the lesion was produced by the serotonin precursor L-5-HTP (5–100 mg/kg i.p.) following peripheral decarboxylase inhibition. Reciprocally, the serotonin releasing agent, PCA<sup>22,29</sup>, produced consistent ipsilateral turning that was blocked by prior depletion of endogenous serotonin stores with PCPA. Several lines of evidence indicate that these effects are at least somewhat specific for increased activity in serotonin-mediated synapses in animals with serotonin nerve terminal destruction. First, no consistent turning in either direction was observed in these animals when administered the catecholamine drugs L-DOPA, apomorphine or D-amphetamine. Second, when L-5-HTP was administered to animals with unilateral 6-OHDA lesions, it never produced consistent turning in either direction, even though these animals turned consistently in response to L-DOPA, amphetamine and apomorphine. Although PCA produced consistent ipsilateral turning in 6-OHDA lesioned animals, the effect was not blocked by prior serotonin depletion. This latter effect is therefore probably attributable to the catecholamine-releasing action of PCA<sup>15,19</sup>. Finally, Hole et al.<sup>12</sup>, utilizing virtually the same technique, i.e., intracerebral injections of 5,7-DHT (4  $\mu$ g base in 4  $\mu$ l saline) in rats pretreated with a catecholamine uptake blocker, recently reported finding little evidence for non-specific damage. There was no change in norepinephrine uptake, and 'no unspecific lesion was observed, except the cannula track and its immediate surroundings (100–150  $\mu$ m).'

Although 5-HTP and PCA produced significant unilateral turning in 5,7-DHT lesioned animals, the magnitude of this effect was considerably smaller than the degree of turning seen in 6-OHDA lesioned animals treated with the various catecholamine drugs. Two hypotheses, which are not mutually exclusive, could account for this difference. First, dopamine, and the neuronal substrates on which it acts, may be more intimately involved in the brain mechanisms subserving the production and stimulation of locomotion or basic motor activities. A large literature exists in support of this basic distinction between the role of dopamine and serotonin in motor output<sup>9,14,17,26,28</sup>. A second, equally appealing, hypothesis relates differences in the absolute amount of turning produced in 5,7-DHT and 6-OHDA animals to differences in unilateral depletion of serotonin and dopamine. A 97% depletion of striatal dopamine and an 85% depletion of forebrain dopamine were found in the 6-OHDA lesioned animals, whereas 5,7-DHT lesioned animals had only 65% and 70% depletion of striatal and forebrain serotonin levels, respectively. Thus, if the degree of depletion produced by injections of 5,7-DHT and 6-OHDA could be more nearly equated, a



greater similarity might be observed in the absolute amount of unilateral turning elicited in animals in these two groups.

The neurochemical data in the present study do not allow us to determine whether destruction of serotonin nerve terminals in the corpus striatum or in the rest of the forebrain is more critical for the manifestation of the unilateral turning phenomenon. As shown in Table II, the mean serotonin depletion in these two areas was approximately equal. Furthermore, a relatively strong correlation existed across individual animals in the amount of depletion found in the two areas, thus making it difficult to attribute the effect to depletion specifically in either locus. It may, of course, be the case that denervation in both areas contributes to the effect in an additive manner. These questions remain to be answered by subsequent experiments employing a more discrete injection technique.

A direct parallel exists between the present results and the results for unilateral 6-OHDA lesioned animals, in that in both cases drugs producing release of endogenous stores of the relevant transmitter result in turning ipsilateral to the lesioned side, whereas drugs having a direct postsynaptic action elicit turning contralateral to the lesioned side. In the case of 6-OHDA lesioned animals, the elicitation of ipsilateral turning by catecholamine releasing agents, such as D-amphetamine<sup>5,11</sup>, has been hypothesized to be due to the increased store of releasable dopamine on the contralateral side<sup>23</sup>. On the other hand, the elicitation of contralateral turning by agents having a direct postsynaptic effect, has been attributed to the greater responsiveness (i.e. supersensitivity) of striatal neurons on the denervated side ipsilateral to the lesion<sup>24,27</sup>. The same arguments may hold for the directionality of turning in 5,7-DHT lesioned animals since we have previously reported that animals administered 5,7-DHT intraventricularly display a supersensitive response to the behavioral effects of L-5-HTP<sup>21</sup>, and a subsensitive response to fenfluramine<sup>21</sup>, a drug which like PCA causes the release of endogenous stores of CNS serotonin<sup>7,22</sup>.

Two recent studies failed to observe consistent unilateral turning in response to L-5-HTP administered to rats with unilateral destruction of forebrain serotonin nerve terminals approximately equivalent in degree to that in the present study<sup>3,16</sup>. In both of these previous cases, however, the level of destruction of dopamine nerve terminals was even greater. One study produced denervation by means of unilateral electrothermal lesions<sup>3</sup>, while the other injected 5,6-DHT directly into the medial forebrain bundle *without* pretreating with a catecholamine uptake blocking agent<sup>16</sup>. It is not clear whether the destruction of dopamine nerve terminals in addition to serotonin nerve terminal destruction in these two studies accounts for the lack of congruence between our results.

In summary, the present results indicate that the turning behavior of rats with unilateral destruction of brain serotonin nerve terminals provides a sensitive tool for quantifiably studying changes in serotonergic function within the central nervous system.

## ACKNOWLEDGEMENTS

This research was supported by Grants MH 23433 and MH 13445 from the National Institute of Mental Health, and the Spencer Foundation.

We would like to thank Merck, Sharpe and Dohme (MK-486), Pfizer (PCPA) and Smith, Kline and French (D-amphetamine) for their generous gift of drugs.

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