BEHAVIORAL AND NEURAL BIOLOGY 46, 491–495 (1986)

BRIEF REPORT

Sex Differences in the Effects of Gonadectomy on Amphetamine-Induced Rotational Behavior in Rats

DIANNE M. CAMP, JILL B. BECKER,[1] AND TERRY E. ROBINSON

Department of Psychology and Neuroscience Laboratory Building, The University of Michigan, Ann Arbor, Michigan 48104-1687

The effects of gonadectomy on amphetamine-induced rotational behavior were studied in male and female rats. Different systemic doses were used to produce equivalent brain concentrations of the drug in each group, thereby controlling for sex differences in the metabolism of amphetamine. Ovariectomy of female rats significantly attenuated amphetamine-induced rotation, whereas castration of males was without effect. The results support the idea that in females, the endogenous gonadal hormones facilitate functional activity in the mesostriatal dopamine system. © 1986 Academic Press, Inc.

It is well established that gonadal steroid hormones influence neural activity in the hypothalamus, a brain area involved in the control of pituitary hormone secretion and reproductive behavior. However, there is growing evidence that functional activity in extrahypothalamic brain areas may also be modulated by gonadal hormones. For example, recent evidence suggests that gonadal steroids modulate activity in the mesostriatal dopamine (DA) system. Several biochemical and behavioral indices of striatal DA activity fluctuate across the estrous cycle (Becker & Ramirez, 1981; Becker, Robinson, & Lorenz, 1982; Robinson, Camp, Jacknow, & Becker, 1982) and are influenced by exogenous hormone treatment (Becker & Beer, 1986). In addition, there are sex differences in a variety of behaviors thought to reflect activity in the mesostriatal DA system, including dopaminergic drug-induced rotation (Robinson, Becker, & Ramirez, 1980; Becker et al., 1982), stereotypy (Beatty & Holzer, 1978), and locomotor activity (Savageau & Beatty, 1981). Females usually show a greater behavioral response to dopaminergic drugs than do males, and

[1] This research was supported by a grant from the NIH (NS16437) to T.E.R. and from the NSF (BNS 84-11763) to J.B.B. Requests for reprints should be sent to Dr. Jill B. Becker, Neuroscience Laboratory Building, The University of Michigan, 1103 E. Huron St., Ann Arbor, MI 48104-1687.

0163-1047/86 $3.00

this may be due to sex differences in circulating levels of gonadal hormones. In support of this, we have reported that ovariectomy (OVX) in females attenuates rotational behavior produced by electrical stimulation of the nigrostriatal DA pathway, but castration (CAST) of males does not (Robinson et al., 1982). The present study was designed to determine if there are similar sex differences in the effects of gonadectomy (GDX) on amphetamine (AMPH)-induced rotational behavior in rats with unilateral 6-hydroxydopamine (6-OHDA) lesions of the nigrostriatal DA system.

Male and female Holtzman rats (Holtzman Co., Madison, WI) were individually housed and maintained on a reverse light/dark cycle (14/10 h, lights off 7:00 AM); food and water were freely available. All animals received an infusion of 8 μg of 6-OHDA · HBr in 4 μl of a 0.1% ascorbic acid–0.9% saline solution into the right substantia nigra. Therefore, 50% of the animals would have a lesion on the "dominant" side for rotational behavior, and 50% on the "nondominant" side (for a discussion see Robinson, Becker, Camp, & Mansour, 1985). Following a recovery period of at least 1 month, animals were either GDX or received a sham operation under ether anesthesia.

Two weeks after GDX rats were tested for AMPH-induced rotational behavior using circular automated rotometers with flat-bottom floors. The rotometers recorded every 90° turn to the right or left, and one rotation was defined as four consecutive 90° turns in the same direction. Following a 15-min habituation period each rat received an ip injection of *d*-AMPH sulfate dissolved in 0.9% saline. Intact females, OVX females, and CAST males received 2.6 mg/kg AMPH, whereas intact males received 3.0 mg/kg. We have previously shown that these doses produce equivalent brain levels of AMPH in these groups (Becker et al., 1982, and unpublished data). After the injection, rats were replaced in the rotometers and rotational behavior was continuously recorded for 2 h. At least 7 days after testing, the animals were killed by decapitation, and the left and right striata were dissected and assayed for DA concentrations using high-performance liquid chromatography with electrochemical detection (Becker & Beer, 1986). To minimize intragroup variation only animals meeting the following criteria were included in the data analyses: (1) the direction of at least 90% of the full rotations made during the 2 h following AMPH was ipsilateral to the lesioned side (only four animals were eliminated due to this criteria alone and they were distributed among three of the groups); and (2) striatal DA depletion was 85% or greater.

The total number of net rotations (i.e., rotations in the preferred direction minus those in the nonpreferred direction) made during the 2 h following the administration of AMPH by intact females (n = 22), OVX females (n = 19), intact males (n = 16), and CAST males (n = 13) are depicted in Fig. 1A. Planned comparisons (one-tailed t tests) were performed to determine whether GDX produced changes in AMPH-induced rotational

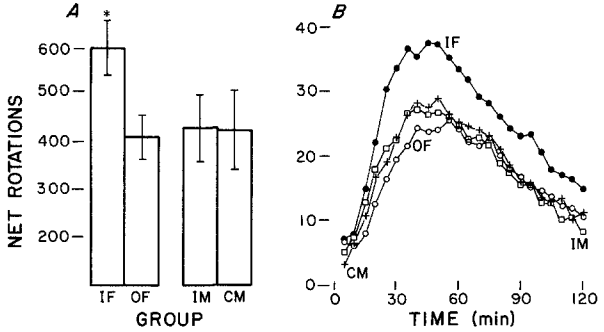

FIG. 1. Amphetamine (AMPH)-stimulated rotational behavior in intact female rats (IF), OVX females (OF), intact males (IM), and CAST males (CM). (A) Cumulative net rotations during the 2 h immediately following AMPH treatment (see text for doses). Bars indicate SEM; * IF > OF (p = .02) and IF > pooled males (p = .027). Male groups did not differ from each other or OF. (B) Time course of rotational behavior over 5-min intervals during the 2-h test session.

behavior that were similar to the changes previously reported for electrical stimulation-induced rotation (Robinson et al., 1982). As predicted, OVX significantly attenuated the turning behavior in females [$t(39) = 2.08$, $p = .02$], whereas CAST had no effect in males [$t(27) = 0.08$]. Because intact and CAST males did not differ, their data were pooled, and the pooled data were then compared to those of intact and OVX females. Males ($n = 29$) made significantly fewer net rotations than intact females [$t(49) = 1.95$, $p = .027$], but did not differ from OVX females [$t(46) = 0.19$].

Figure 1B shows that the effect of OVX on rotational behavior is primarily due to a decrease in the number of net rotations made during the first hour following AMPH administration. Analyses of variance with repeated measures on each 5-min interval during the first 60 min of rotational behavior produced significant interaction effects for intact and OVX females [$F(11, 429) = 3.38$, $p < .001$], and intact females and pooled males [$F(11, 539) = 2.33$, $p = .008$]. Both interactions are due primarily to the greater increase in net rotations made by intact females beginning 15 to 20 min following AMPH administration. Although intact females continued to display more rotational behavior than OVX females, intact males, and CAST males during the second 60 min following AMPH, these differences were not statistically significant. Finally, OVX females, intact males, and CAST males did not differ from each other in their pattern of rotational behavior over the first or second hour following AMPH administration.

The present study supports previous reports of sex differences in AMPH-induced rotational behavior (Robinson et al., 1980; Becker et al., 1982).

Intact females made significantly more net rotations in response to an acute injection of AMPH than intact males. In addition, OVX attenuated AMPH-stimulated rotational behavior and effectively abolished the sex difference in rotational behavior. In contrast, CAST of males had no observable effects on AMPH-induced rotational behavior. These latter results strongly suggest that the sex difference in AMPH-induced rotational behavior is due to sex differences in circulating levels of ovarian hormones.

It is unlikely that the present results are due to sex differences in the metabolism of AMPH. AMPH is metabolized more rapidly in male rats than in females. Therefore, if males and females are given the same systemic dose of AMPH, brain concentrations are greater in females (Becker et al., 1982). However, in the present study we controlled for this variable by using systemic doses of AMPH that produce equivalent brain levels of AMPH in unlesioned male and female rats (Becker et al., 1982). Although rats with 6-ODHA lesions were used here, and it is known that this damages the blood–brain barrier, by 21 days postlesion the blood–brain barrier is restored (Cooper, Novin, & Butcher, 1982). The doses of AMPH used here should produce brain concentrations of AMPH comparable to those found in intact animals because rats were not tested until at least 6 weeks postlesion, allowing plenty of time for recovery of the blood–brain barrier. In addition, it is unlikely that the effects reported here are due to some kind of unique interaction between AMPH and circulating gonadal hormones, as rotational behavior produced by electrical stimulation of the nigrostriatal DA pathway is also attenuated following OVX but not after CAST (Robinson et al., 1982).

The findings are consistent with a growing body of literature suggesting that gonadal hormones can influence functional activity in the mesostriatal DA system. However, the ovarian hormones appear to play a more important role in the modulation of presynaptic mesostriatal DA activity than do male gonadal hormones. If it is assumed that a decrease in AMPH-elicited rotational behavior reflects a decrease in mesostriatal DA activity, then ovarian hormones facilitate the presynaptic activity of mesostriatal dopaminergic neurons. This idea is supported by experiments showing that OVX attenuates the AMPH-stimulated release of striatal DA, and that estrogen or estrogen plus progesterone replacement therapy restores striatal DA release comparable to that of intact females (Becker & Ramirez, 1981; Becker et al., 1984; Becker & Beer, 1986). In contrast, CAST of male rats has no effect on AMPH-stimulated striatal DA release (Becker & Ramirez, 1981). One would expect that other behaviors dependent on mesostriatal DA release would also be attenuated following OVX. However, one cannot assume that all behaviors induced by dopaminergic drugs will be similarly affected. There are probably both presynaptic and postsynaptic changes in the mesostriatal DA system associated with ovarian hormones, and drugs that act on DA receptors

may produce very different results from the effect of AMPH (for discussion see Becker & Beer, 1986).

In closing, it is worth noting that these findings may have clinical relevance. For example, choreiform disorders that are thought to be due to hyperactive DA systems have been reported as a side effect of the contraceptive pill and during pregnancy, and both conditions are associated with elevated levels of ovarian hormones (for discussion see Robinson et al, 1980). Therefore, further research on how sex and gonadal hormones influence functional activity in the mesostriatal DA system may provide a better understanding of the factors that precipitate clinical disorders related to DA dysfunction and may be useful in formulating alternate methods of treatment.

REFERENCES

Beatty, W. W., & Holzer, G. A. (1978). Sex differences in stereotyped behavior in the rat. *Pharmacology, Biochemistry and Behavior,* **9,** 777–783.

Becker, J. B., & Beer, M. E. (1986). The influence of estrogen on nigrostriatal dopamine activity: Behavioral and neurochemical evidence for both pre- and postsynaptic components. *Behavioural Brain Research,* **19,** 27–33.

Becker, J. B., Beer, M. E., & Robinson, T. E. (1984). Striatal dopamine release stimulated by amphetamine or potassium: Influence of ovarian hormones and the light–dark cycle. *Brain Research,* **311,** 157–160.

Becker, J. B., & Ramirez, V. D. (1981). Sex differences in the amphetamine-stimulated release of catecholamines from rat striatal tissue in vitro. *Brain Research,* **204,** 361–372.

Becker, J. B., Robinson, T. E., & Lorenz, K. A. (1982). Sex differences and estrous cycle variations in amphetamine-elicited rotational behavior. *European Journal of Pharmacology,* **80,** 65–72.

Cooper, P. H., Novin, D., & Butcher, L. L. (1982). Intracerebral 6-hydroxydopamine produces extensive damage to the blood–brain barrier in rats. *Neuroscience Letters,* **30,** 13–18.

Robinson, T. E., Becker, T. E., Camp, D. M., & Mansour, A. (1985). Variation in the pattern of behavioral and brain asymmetries due to sex differences. In S. D. Glick (Ed.), *Cerebral lateralization in nonhuman species* (pp. 185–231). New York: Academic Press.

Robinson, T. E., Becker, J. B., & Ramirez, V. D. (1980). Sex differences in amphetamine-elicited rotational behavior and the lateralization of striatal dopamine in rats. *Brain Research Bulletin,* **5,** 539–545.

Robinson, T. E., Camp, D. M., Jacknow, D. S., & Becker, J. B. (1982). Sex differences and estrous cycle dependent variation in rotational behavior elicited by electrical stimulation of the mesostriatal dopamine system. *Behavioural Brain Research,* **6,** 273–287.

Savageau, M. M., & Beatty, W. W. (1981).Gonadectomy and sex differences in the behavioral response to amphetamine and apomorphine. *Pharmacology, Biochemistry and Behavior,* **14,** 17–21.