*Pharmacology Biochemistry & Behavior*, Vol. 18, pp. 285–287, 1983. Printed in the U.S.A.

BRIEF COMMUNICATION

# Open-Field Behavior in Dopamine-Depleted Rat Pups and Their Mothers

JAMES T. CONCANNON AND MARTIN D. SCHECHTER

*Department of Pharmacology, Northeastern Ohio Universities College of Medicine, Rootstown, OH*

Received 19 April 1982

CONCANNON, J. T. AND M. D. SCHECHTER. *Open-field behavior in dopamine-depleted rat pups and their mothers.* PHARMACOL BIOCHEM BEHAV **18**(2) 285–287, 1983.—Possible changes in the behavior of rat mothers and their pups were investigated by administering intracisternal injections of 6-hydroxydopamine (6-OHDA) or its vehicle to 5-day-old rats. Administration of the neurotoxin resulted in a significant depletion of whole-brain dopamine levels to 23% of control levels, whereas norepinephrine levels were reduced to 83% of controls. Open-field behavior revealed that the 6-OHDA-treated rat pups were hypoactive, in terms of decreased square crossings, at 15 days of age, yet were hyperactive at 30 days of age. Toxin-treated pups also showed lower urination scores at 25 and 30 days of age. Mothers' open-field behavior was virtually unaffected by the treatment status of their offspring (i.e., 6-OHDA vs. vehicle-treated), although several of the mothers' behaviors decreased with repeated measures over days.

Hyperactivity Attentional deficit disorder 6-Hydroxydopamine Activity Developing rats Open-field behavior Animal models Emotionality

ALTHOUGH neonatal 6-hydroxydopamine administration reliably produces both dopamine depletion and hyperactivity in developing rat pups [2,9], little is known concerning possible interactions between hyperactive rat pups and their mothers [6,7]. For example, it is possible that mothers of hyperactive pups may be more "emotional" [2] in open-field tests, or may respond in some other way to sustain pup hyperactivity (c.f. [5]). On the other hand, the presence of the mother may suppress hyperactivity which is most readily seen when dopamine-depleted pups are tested in isolation [9]. Hence, the present study was designed to examine possible mother-pup interactions during the course of developmental hyperactivity.

## METHOD

### *Animals*

Male and female offspring of Sprague-Dawley-derived rats, born and raised in the Department colony in litters culled to 8 pups, served as subjects. Animals were housed under controlled temperature and a 12-hour light/12-hour dark cycle. Food and water were provided ad lib.

### *Apparatus*

The open-field consisted of a 1.0×1.0×0.3 m topless rectangular painted wooden box, with a thin plastic floor divided into 100 10×10 cm squares. Placed in the center of this arena was a 26×26×26 cm clear Plexiglas box raised from the floor by approximately 1 cm.

### *Procedure*

At 5 days of age, litters of rat pups were randomly assigned to one of two treatment groups: (a) intraperitoneal (IP) desmethylimipramine (30 mg/kg:1 ml/kg) and intracisternal (IC) 6-OHDA (150 μg in 25 μl of distilled water with 0.4 mg ascorbate/ml) or (b) IP DMI and IC vehicle [1,8]. All animals within a litter were treated identically (i.e., with 6-OHDA or vehicle).

Open-field behavior was then determined for the mothers and pups at 15, 20, 25 and 30 days of age. Pups were placed within the Plexiglas enclosure while the mothers remained outside of it. Animals were placed in the apparatus for 3 min, and the number of square crossings (horizontal activity), assisted (climbing) and non-assisted rearings (vertical activity), fecal boli, and the presence or absence of urination were recorded. Within each day, the same mother interacted with 2 male and 2 female pups given the same brain treatment using a 60-min intertrial interval. All observations were collected between 1300 and 1600 hours. At 35 days of age, all pups were sacrificed by decapitation and their brains were analyzed for levels of dopamine (DA) and norepinephrine (NE) using previously-described methods [1,3].

### *Statistical Methods*

Horizontal activity, vertical activity, defecation, and urination of the pups were examined using 4 separate 2 (6-OHDA vs. vehicle) × 4 (age) mixed analyses of variance (ANOVAs), with age representing the repeated measure.

TABLE 1

SUMMARY OF PUPS' BEHAVIOR AND BODY WEIGHTS

| Age | Treatment: Vehicle | Treatment: Toxin | Age | Treatment: Vehicle | Treatment: Toxin |
|---|---|---|---|---|---|
| | Panel A: Horizontal Activity | | | Panel B: Vertical Activity | |
| 15 | 64.93 ± 6.44 | *46.09 ± 6.12 | 15 | 8.00 ± 1.29 | *4.91 ± 0.65 |
| 20 | 71.61 ± 5.95 | 65.81 ± 4.72 | 20 | 11.95 ± 1.28 | *8.44 ± 0.73 |
| 25 | 56.64 ± 5.60 | 63.28 ± 6.68 | 25 | 12.25 ± 1.26 | 10.09 ± 1.10 |
| 30 | 35.75 ± 4.34 | *53.06 ± 7.31 | 30 | 6.18 ± 1.00 | *9.47 ± 1.06 |
| | Panel C: Defecation Score | | | Panel D: Urination Score | |
| 15 | 0.18 ± 0.13 | 0.06 ± 0.06 | 15 | 0.14 ± 0.07 | 0.06 ± 0.04 |
| 20 | 0.82 ± 0.21 | 0.25 ± 0.13 | 20 | 0.25 ± 0.08 | 0.06 ± 0.04 |
| 25 | 1.32 ± 0.33 | 0.87 ± 0.27 | 25 | 0.39 ± 0.09 | *0.19 ± 0.07 |
| 30 | 1.68 ± 0.40 | 1.06 ± 0.28 | 30 | 0.32 ± 0.09 | *0.12 ± 0.06 |
| | Panel E: Body Weights (grams) | | | | |
| 5 | 11.20 ± 0.25 | 11.11 ± 0.35 | | | |
| 15 | 26.25 ± 0.61 | *23.78 ± 0.87 | | | |
| 20 | 36.32 ± 0.67 | 33.83 ± 1.32 | | | |
| 25 | 55.46 ± 1.17 | *47.14 ± 2.15 | | | |
| 30 | 78.03 ± 1.67 | *63.20 ± 3.10 | | | |
| 35 | 101.11 ± 2.21 | *82.81 ± 4.17 | | | |

All values are Mean ± S.E.M.
*$p<0.05$ using Duncan's Multiple Range Test.

Subsequent to the ANOVAs, all between-group comparisons were made utilizing Duncan's Multiple Range Test. Similar ANOVAs were conducted for mothers' behavior using each mother-child "interaction" as the unit of analysis. Throughout the experiment $p<0.05$ was considered to be statistically significant.

## RESULTS

The horizontal activity measure revealed that the 6-OHDA-treated pups were hypoactive at 15 days of age (Table 1, Panel A), but were hyperactive at 30 days of age. Similar results were seen for vertical activity (Table 1, Panel B). Pups did not differ in defecation rates (Table 1, Panel C), although urination rates (Table 1, Panel D) were lower in the 6-OHDA-treated rats at 25 and 30 days of age.

Hyperactivity was not due solely to body weight differences [4] (Table 1, Panel E), since 6-OHDA-treated pups weighed less than normal rats at both 15 and 30 days of age, although they were hypoactive at 15 days of age, but hyperactive at 30 days of age. Furthermore, there was no striking correlation between activity and body weight at 30 days of age (r's = −0.31 and 0.11 respectively, for horizontal and vertical activity) when 6-OHDA-treated pups were hyperactive.

Mother's behavior (data not shown) revealed few systematic between-group differences at any observation time. However, most of the indices showed a decrease over days. Correlations between mothers' and pups' behavior were not calculated at any age due to lack of between-group differences in mothers' behavior.

Mean (±S.E.M.) whole-brain DA concentrations in pups were 177.86±30.20 ng/g and 785.46±11.45 ng/g for the 6-OHDA and normal pups, respectively. This represents a 77.4% depletion which was statistically significant. Whole-brain NE levels were 270.54±12.52 ng/g and 327.20±4.96 ng/g for the 6-OHDA- and vehicle-treated pups, respectively.

## DISCUSSION

The results of this study indicate that neonatal 6-OHDA administration, although producing developmental hyperactivity in DA-depleted rat pups, does not alter mother-child interactions at the ages examined. To the best of our knowledge, this represents the first report of 6-OHDA-induced hyperactivity in the open-field situation, and shows that 6-OHDA-treated pups are hyperactive in situations other than isolation [9]. Failure of pup status to alter mothers' activity was previously reported by Piccirillo *et al.* [7] for pups 3–13 days of age, although pup locomotor activity was not measured in this study.

Failure to find an alteration in mothers' behavior as a function of the pups' hyperactivity might be explained in at least two ways. First, hyperemotionality of rat mothers may have arisen had the experimental sessions been longer, although they were long enough to reveal between-group differences in the pups. Second, mothers' hyperemotionality might have been detected had we examined mother-child interactions at ages earlier than 15 days of age (cf. [7]), although it must be remembered that 6-OHDA-induced hyperactivity in the pups does not occur before 15 days of age.

## ACKNOWLEDGMENTS

The authors would like to thank Monica Budac for her technical assistance and Patricia McGinley and Denise Lovano for assistance in brain assays. Supported in part by Public Health Service grant MH-33636.

## REFERENCES


1. Concannon, J. T. and M. D. Schechter. Hyperactivity in developing rats: Sex differences in 6-hydroxydopamine and amphetamine effects. *Pharmacol Biochem Behav* **14:** 5–10, 1981.
2. Denenberg, V. H. Open-field behavior in the rat: What does it mean? *Ann NY Acad Science* **159:** 852–859, 1969.
3. Felice, F. J., J. D. Felice and P. T. Kissinger. Determination of catecholamines in rat brain parts by reverse-phase ion-pair liquid chromotography. *J Neurochem* **31:** 1461–1465, 1978.
4. Loch, R. K., L. S. Rafales, I. A. Michaelson and R. L. Bornschein. The role of undernutrition in animal models of hyperactivity. *Life Sci.* 22: 1963–1970, 1978.
5. Ney, P. G. Four types of hyperkinesis. *Can J Psychiatr* **19:** 543–550, 1974.
6. Piccirillo, M., J. E. Alpert, D. J. Cohen and B. A. Shaywitz. Effects of 6-hydroxydopamine and amphetamine on rat mothering behavior and offspring development. *Pharmacol Biochem Behav* **13:** 391–395, 1980.
7. Piccirillo, M., D. J. Cohen, B. A. Shaywitz, J. E. Alpert and D. Marinelli. Maternal care received by rat pus treated with 6-hydroxydopamine. *Physiol Behav* **22:** 69–75, 1979.
8. Shaywitz, B. A., R. D. Yager and J. H. Klopper. Selective brain dopamine depletion in developing rats: An experimental model of minimal brain dysfunction. *Science* **191:** 305–308, 1976.
9. Teicher, M. H., B. A. Shaywitz, H. L. Kootz and D. J. Cohen. Differential effects of maternal and sibling presence on hyperactivity of 6-hydroxydopamine-treated developing rats. *J Comp Physiol Psychol* **95:** 134–145, 1981.