Pharmacology Biochemistry & Behavior, Vol. 19, pp. 743–749, 1983.

Differential Effects of Selective Dopamine, Norepinephrine or Catecholamine Depletion on Activity and Learning in the Developing Rat

LISA A. RASKIN[1]

Department of Psychology, Amherst College, Amherst, MA 01002

BENNETT A. SHAYWITZ, GEORGE M. ANDERSON, DONALD J. COHEN, MARTIN H. TEICHER[2] AND JAMES LINAKIS

Yale University School of Medicine, 333 Cedar Street, New Haven, CT 06510

Received 27 May 1983

RASKIN, L. A., B. A. SHAYWITZ, G. M. ANDERSON, D. J. COHEN, M. H. TEICHER AND J. LINAKIS. *Differential effects of selective dopamine, norepinephrine or catecholamine depletion on activity and learning in the developing rat.* PHARMACOL BIOCHEM BEHAV **19**(5) 743–749, 1983.—The present experiment investigated the behavioral changes which occurred following neonatal depletion of central catecholamine systems in the rat. The behavioral effects which resulted from selective dopamine (DA) depletion were compared with those resulting from selective norepinephrine (NE) depletion as well as depletion of both catecholamines (CA). Neonatal 6-hydroxydopamine (6-OHDA) was administered intracisternally at 5 days of age following pretreatment with desmethylimipramine in order to selectively deplete DA. NE levels were reduced by intraperitoneal injections of 6-OHDA at 1 and 2 days of age. Depletion of both catecholamines was effected by combining the procedures used for selective depletion of both DA and NE. Activity was time sampled during an hour at 3 preweanling ages. Avoidance and escape learning were measured in a T maze when pups were 20 days of age and in a Shuttlebox apparatus on day 28. Results revealed that DA and CA depleted animals were hyperactive in comparison to controls and displayed severe learning impairments in both T maze and Shuttlebox performance. In contrast, NE depleted animals showed activity levels which were similar to controls but were significantly impaired on both learning paradigms. These results suggest that selective lesions of DA and NE in infancy lead to a constellation of behaviors which are distinctly unique. The implications of these findings is discussed in terms of clinical research into the Attentional Deficit Disorder of childhood.

Catecholamines 6-OHDA Neonatal Activity

THERE is a great deal of evidence which suggests that early catecholamine (CA) systems influence activity [4, 5, 9, 15] and learning [15,23] during development in the rat. Neonatal lesions of central CA systems with the neurotoxin 6-hydroxydopamine (6-OHDA) produces rats that are hyperactive [9, 14, 15] and show severe learning impairments [14] throughout ontogeny. Although the behavioral deficits which result from early CA depletion have been substantiated the contributory role that each CA system plays in producing these behavioral deficits is less clear.

Selective neonatal dopamine (DA) depletion with 6-OHDA and desmethylimipramine (DMI) has consistently been reported to produce hyperactivity during development [7, 15, 25]. The magnitude of activity reportedly depends upon the extent of the lesion [13] and the age at which it is effected [7]. In addition, neonatal DA lesions lead to severe learning impairments, which are apparently independent of the hyperactivity-inducing effect of the lesion. Weldon *et al.* [28] report that learning deficits can be observed in neonatally DA depleted animals several days before lesion-induced hyperactivity begins to emerge.

Reports concerning the role that developing norepinephrine (NE) systems play in the ontogeny of learning and activity have been inconsistent. The extent to which early destruction of NE systems leads to behavioral impairments appears to depend upon the route of administration of 6-OHDA, the CNS depletion levels, as well as methodologies employed. For example Pappas *et al.* [16] have

[1]The preparation of this manuscript was supported by an Amherst College Trustee Faculty Fellowship.

[2]Now at Department of Psychiatry, Harvard Medical School, McLean Hospital, Mailman Research Laboratories, Belmont, MA 02178.

reported no change in behavior following subcutaneous neonatal 6-OHDA injections; a treatment which depleted peripheral as well as cortical NE. Although depletion levels were considerable in this study, activity was measured for only 5 minutes and learning tasks consisted of a criterion of only 3 consecutive shock avoidances. Thus, the behavioral observations in this experiment were relatively limited. In another experiment, Shaywitz *et al.* [24] also reported no change in either activity or learning following neonatal intracisternal 6-OHDA. Although in this experiment activity was observed throughout a 60 minute session, and two learning paradigms were utilized, each with a rigorous acquisition period, the depletion schedule was not very selective for NE. Using this protocol a 40% reduction in whole brain NE was accomplished along with a 20% reduction in whole brain DA. Other experiments in which forebrain NE was selectively depleted by neonatal intraperitoneal (IP) injections of 6-OHDA [14] or 6-OHDOPA [12] produced animals with severe learning impairment and hyperactivity, however, locomotor activity was only observed in these latter experiments during a 15 minute session. Most recently Shaywitz *et al.* [23] injected neonatal 6-OHDA IP which produced a significant depletion in forebrain NE. Activity was time-sampled throughout the course of an hour and animals were tested on both T-maze and Shuttlebox avoidance paradigms. The results of this report showed that early NE depletion led to severe learning impairments without any change in activity during ontogeny.

Thus, an accumulation of the existing evidence suggests that early lesions of DA and CA systems produce hyperactivity and learning impairments in rats [7, 9, 15, 25]. Animals with lesions of NE systems show learning impairments without hyperactivity, when activity is observed for at least an hour, acquisition schedules for learning paradigms are rigorous, and the depletion protocol is selective for NE [23].

In most previous experiments, an attempt has been made to isolate the influence of early damage to one or another catecholamine system on behavior. Given that the methodology of experiments varies considerably between laboratories, the purpose of the present experiment was to directly compare the influence of early DA, NE and CA depletion on the development of activity and learning in the rat.

METHOD

Subjects

The subjects were 60 male and female Sprague-Dawley rats born to 6 litters, which were obtained from Charles River Laboratories when the pups were 1 day of age. Litters were culled to 10 pups, at day 1, and placed in clear plastic cages (30×32×10 cm) with pine shavings on the floor. All animals were housed in a room with a 12 hour light (lights on 0700) and 12 hour dark diurnal cycle which was maintained at 21°C. A split litter design was employed so that no more than 3 animals from each litter were assigned to a treatment group. All pups were randomly assigned to one of the following experimental conditions: Preferential NE depletion, control for NE depletion, preferential DA depletion; control for DA depletion, catecholamine depletion and the appropriate control. All pups were weighed weekly throughout the course of the experiment.

Procedure

Pharmacological preparations. 6-Hydroxydopamine, obtained from Regis Chemical Company in Chicago was prepared immediately before injections in 0.9% isotonic saline solution containing 0.4 mg/ml of ascorbic acid. Control solutions consisted of 0.9% isotonic saline and ascorbic acid.

Selective NE depletion was effected by injecting 100 mg/kg of 6-OHDA IP at 1 and 2 days of age, a procedure shown to selectively deplete forebrain NE [3]. Control animals for this group were given the vehicle solution IP at days 1 and 2. Preferential depletion of brain DA was accomplished by procedures which have previously been reported [24,25]. At 5 days of age, pups were pretreated with 20 mg/kg of desmethylimipramine (DMI, USV Pharmaceuticals) administered IP which was followed an hour later, by an intracisternal (IC) injection of 100 μg of 6-OHDA (free base) in 20 μl of solution. Intracisternal injection was performed by flexing the neck of the infant rat and injecting the solution via a precalibrated Hamilton microsyringe with a No. 27 gauge needle which was inserted below the occiput. Control animals received the vehicle solution injected IC at 5 days of age. Both catecholamines were depleted by utilizing the procedure which depletes DA as well as that which depletes NE. Consequently, pups were injected IP at day 1 and 2 with 6-OHDA and at 5 days of age were given IC injections of 6-OHDA following DMI pretreatment. Control animals for this treatment group received IP injections on day 1 and 2 and IC injections on day 5 with saline and ascorbic acid.

Activity

Motor activity was observed when the pups were 12, 19 and 26 days of age. Each pup was placed alone in a plastic cage (20×50×15 cm) which had been painted with nonreflecting black paint. All animals were given free access to food and water. The activity cages were placed in a soundproof room (2.5×4 meters) which was illuminated by six 150 W infra red heat lamps (General Electric) and was maintained at 25°C. Activity was observed for a 1 hour period via a video camera (Panasonic, Model No. WV-261) mounted above the cages, which was connected to an Hitachi time-lapse tape recorder (Model No. 6M-912). Tapes were played back and scored at a speed which was 6 times faster than recorded speed and an activity score was taken for every other 5 minute period throughout the hour, beginning with the first 5 minutes of the hour. Activity was scored as the duration of time each animal spent engaged in any movement, such as, walking, running, twitching, rearing, grooming, etc.

T Maze Escape

Escape performance in a T maze apparatus was measured at 20 days of age [24,25]. In this paradigm, each animal was placed in an opaque Plexiglas T maze with a grid floor. The long arm of the maze was 31 cm long and each cross piece was 16 cm long. Shock was administered by activating the grid with a 2 mA current via a shock scrambler (BRS/LVE Model No. SGS-004). The animal was placed on the grid in the long end of the T maze and was required to run to a cross piece of the maze in order to avoid shock. Each session began when the experimenter lifted a starting gate which activated the shock scrambler, and was terminated when the animal entered the safe compartment. Entry into a safe compartment was recorded by a light switch (Sigma Instrumental 8RCOIA) which was activated by the animal's presence. Each animal was given 5 blocks of trials, with each block consisting of 4 trials. The latency to escape from the long

arm into a safe compartment was recorded. From those data, the number of trials in which the animal did not escape within 30 seconds was determined.

Shuttlebox Escape and Avoidance

Escape and avoidance performance was measured at 28 days in the manner which has also been previously reported [22, 23, 24]. The animal was placed in a Plexiglas Shuttlebox with stainless steel rods on the floor. Each of the two compartments were 20×14×17 cm and were separated by a 5 cm high wall. The starting compartment was painted black and the goal compartment was white and was illuminated by a flashlight bulb.

Each animal was trained by being placed into one compartment and sounding a bell for 1 sec. This was followed 5 seconds later by the 2.5 mA shock, which was administered via the shock scrambler to the stainless steel rods. If the animal did not escape into the safe compartment with 30 sec, he was placed there by the experimenter for 20 sec. When the animal entered the safe compartment a light switch was activated which stopped the timer (Sigma Instruments, Model B8RCOIA). The latency to avoid the shock as well as the number of failures to avoid shock was recorded for every animal. Each animal was given 5 blocks of trials with each trial consisting of 4 escape sessions.

Biochemical Analysis

The rats were sacrificed by decapitation and the whole brain was placed on an ice cooled petri dish. Each brain was dissected into the following sections: frontal cortex, striatum, brainstem and hypothalamus. Frozen brains were stored at −70°C and dopamine, norepinephrine and both catecholamines were assayed with the high performance liquid chromatography method (HPLC) [2].

RESULTS

A few animals from each treatment group did not survive to be tested on the behavioral measures. Consequently, the data analyses for activity and T maze measures were performed for the following numbers of SS: NE depleted n=10, DA depleted n=7, CA depleted n=8 and controls n=25. For the Shuttlebox paradigm the number of subjects in the treatment groups were the same; however, one control animal died prior to testing resulting in n=24. Preliminary analyses were performed for all behaviors comparing the different control groups and given that no differences emerged the results from all the control groups were pooled. In addition, the body weights for animals in all groups were not significantly different, $p<0.05$.

Biochemical Analysis

The results of the HPLC assays are shown in Fig. 1. This figure illustrates the catecholamine concentrations in each of the four brain regions expressed as a percentage of control values. Bars above the line indicate that the treated group had mean concentrations greater than controls, while bars below the line indicate a reduction in the level of catecholamine in comparison with controls. The NE depleted group showed a 55% reduction in frontal cortex NE, accompanied by a 45% increase from control values in noradrenergic activity in the brain stem. No change from control values was observed in either the striatum or the hypothalamus. The DA depletion schedule produce approximately a 96% reduction in DA level in the striatum, but no significant reduction in any other brain region. CA levels in the CA depleted animals showed an 80% reduction in forebrain norepinephrine as well as a 98% reduction in dopamine in the striatum and a 60% reduction in hypothalamic DA. One way analysis of variance (ANOVA) and Dunnett's one tailed t tests were performed to assess the extent of depletion on each brain region in all four treatment groups. Although biochemical analyses were performed on the brains from all animals used in the behavioral testing, occasional technical problems with the HPLC assay account for the fact that data from less than 50 animals were reported.

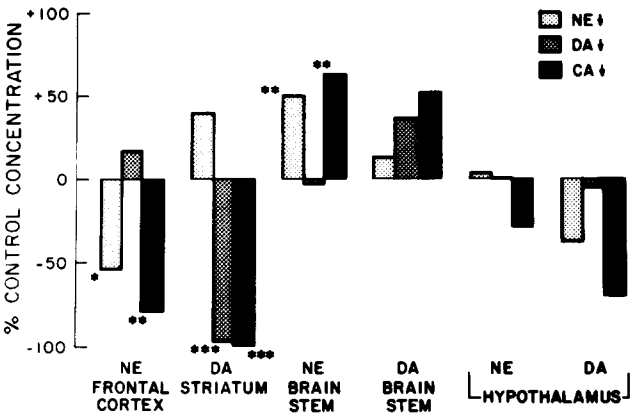

FIG. 1. Concentrations of norepinephrine (NE) and dopamine (DA) and catecholamines (CA) in frontal cortex, striatum, brain stem and hypothalamus. Values are plotted as percentage of that observed in controls, (*$p<0.05$, **$p<0.01$, ***$p<0.001$).

An ANOVA on NE levels in the cortex showed a significant effect of treatment, $F(3,36)=8.48, p<0.001$. Post hoc t's revealed that NE and CA depletion produced NE levels in the cortex which were significantly lower than controls ($p<0.05$, $p<0.01$ respectively), yet DA depletion did not significantly affect NE in the cortex ($p>0.05$). NE in the brainstem was significantly affected by treatment, $F(3,42)=18.87$, $p<0.001$, for both the NE and CA depleted animals ($p<0.01$) but not for DA depleted animals ($p>0.05$). DA levels in the striatum were significantly reduced, $F(3,38)=24.96$, $p<0.001$, for the DA and CA depleted animals ($p<0.001, p<0.001$) but were not significant for NE depleted subjects, $p>0.05$. Treatment appeared to produce catecholamine depletion in the hypothalamus, however, none of these differences were significant due to extreme variability.

Activity

The results of neurochemical treatment on the percentage of time that animals were active, are shown in Fig. 2. Activity levels changed during development for animals in all treatment groups. There was a significant effect of age, but no age × treatment interaction and to facilitate comparisons activity levels were combined for all age groups. Activity is plotted as a function of 6 trials, each trial consisting of alternating 5 minute intervals throughout the hour. Trial I constitutes the first 5 minutes of the hour. Fig. 2 shows that control animals were active when they were first placed into a novel cage, however, their activity subsided as they became accli-

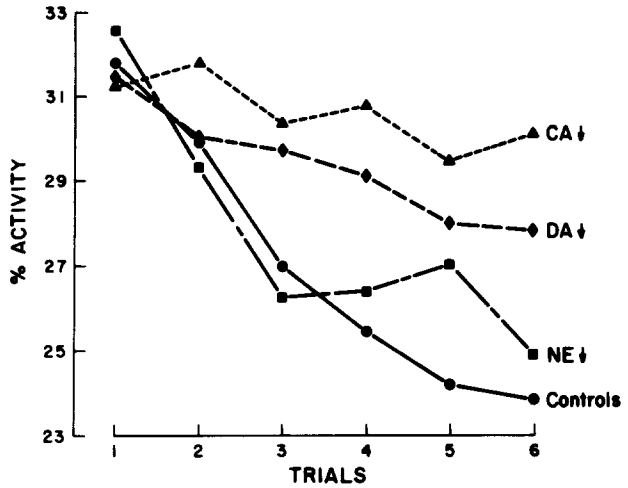

FIG. 2. Percentage time spent active plotted as a function of alternating 5 minute periods for NE, DA, CA depleted animals and controls. Values represent the means for all age groups tested.

mated to the environment. The NE depleted animals exhibited an activity pattern which closely paralleled that observed in controls. In contrast, both the DA and CA depleted animals remained hyperactive throughout the test session. A two-way ANOVA confirmed this pattern of results and showed an effect of neurochemical treatment, $F(3,46)=3.8$, $p<0.05$; an effect of time period, $F(5,46)=14.65$, $p<0.001$ and a treatment × time interaction, $F(15,230)=1.77$, $p<0.05$. A subsequent two-way ANOVA comparing activity for NE depleted animals with that of controls over time revealed no difference in activity between these two groups, $F(1,33)=0.472$, $p>0.05$. In this analysis, time was a significant factor, $F(5,33)=20.37$, but there was no treatment × time interaction, $p>0.05$. Thus hyperactivity, as well as a failure to habituate to the test cage, was observed in DA and CA depleted groups. NE depleted animals responded similarly to control animals both in level of activity and habituation to the test chamber.

T Maze Performance

Escape performance in a T maze at 20 days of age is shown in Figs. 3 and 4. Figure 3 shows the latency to escape shock for controls and treated animals. Clearly control animals escape much more quickly than all treated groups which do not appear different from each other. A one way ANOVA confirmed an overall effect of treatment, $F(3,46)=3.43$, $p<0.05$. Figure 4 shows the percentage of animals in each experimental group who failed to escape the shock within a 30 second period collapsed over the 5 time periods. Again, it is clear that NE, DA and CA depleted animals showed deficits in learning in that significantly more animals in these groups than in control groups failed to escape shock within a 30 second period. Analysis of variance confirmed an effect of treatment, $F(3,46)=3.06$, $p<0.05$. Post hoc t tests showed that the NE, DA, and CA groups were significantly different than controls but not different from each other, $p<0.05$.

Shuttlebox Performance

Shock avoidance examined at 28 days in a one way

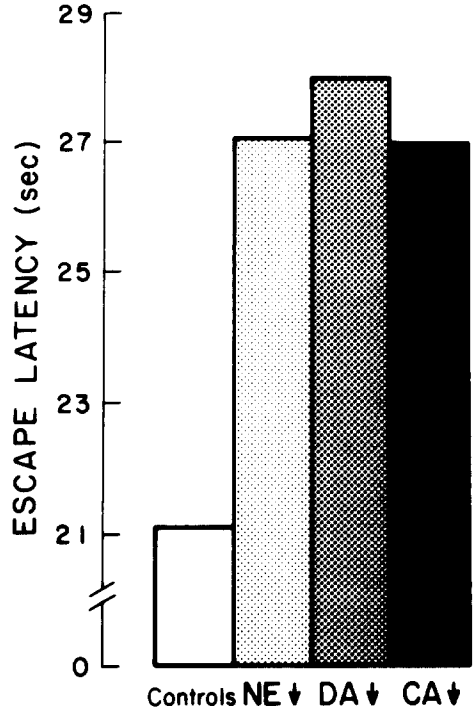

FIG. 3. Latency to escape from shock in the T maze apparatus for NE, DA, CA depleted and control animals.

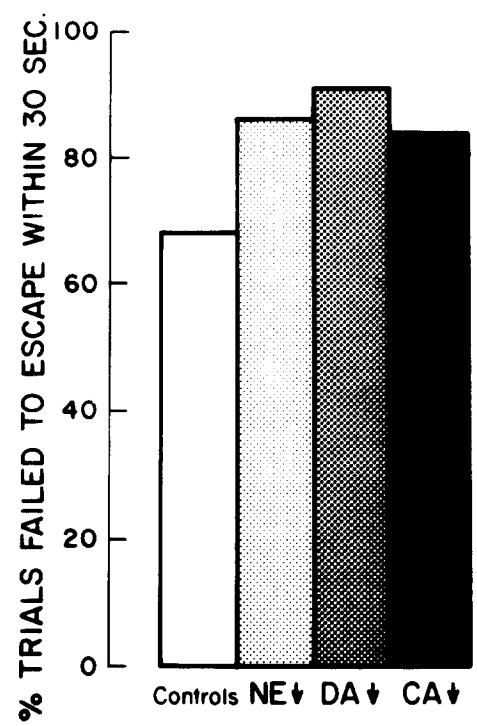

FIG. 4. Percentage of trials on which animals failed to avoid shock in the T maze within 30 seconds for NE, DA, CA depleted animals and control values. Control values were significantly different than those for all treated groups. Treated groups did not differ from each other.

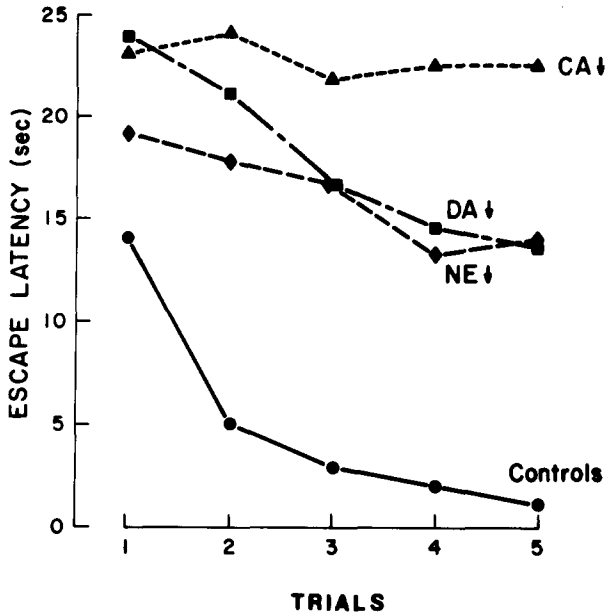

FIG. 5. Latency to escape shuttlebox apparatus plotted as a function of blocks of trials.

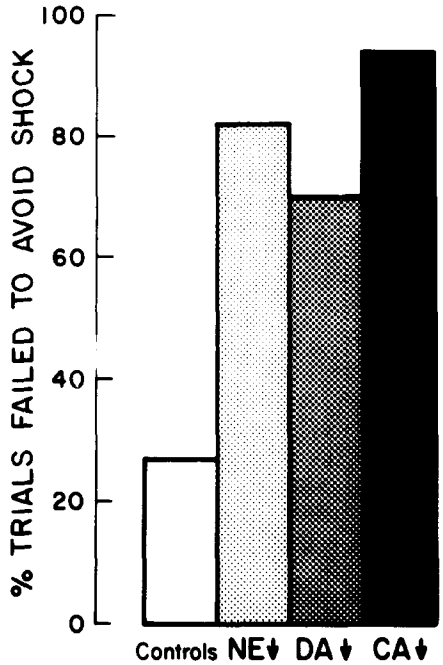

FIG. 6. Percentage of trials on which animals failed to avoid shock in Shuttlebox within 6 seconds after a warning signal for NE, DA, CA depleted and control animals. Control animals were significantly different than all other groups. Treatment groups were not statistically different from each other.

shuttlebox is shown in Figs. 5 and 6. Figure 5 shows the escape latency plotted as a function of time for treatment and control groups. Control animals showed a decrease in escape latency after the first trial and very short escape latencies throughout the test session. In contrast, NE and DA depletion animals demonstrated significantly longer escape latencies. Animals depleted of both catecholamines showed the longest latency to escape and did not improve performance throughout the test session. A treatment × trials two way ANOVA with one repeated measure revealed a significant effect of treatment, $F(3,34)=23.15$, $p<0.001$, a significant trials effect, $F(4,180)=27.69$, $p<0.001$, and a treatment × trials interaction, $F(12,180)=4.35$, $p<0.001$. A subsequent analysis showed that DA and NE depleted animals did not differ from each other and that both groups showed a decrease in escape latency over time; $F(1,15)=0.54$, $p<0.05$ for treatment, $F(4,60)=7.63$, $p<0.05$ for trials and no treatment by trials interaction. Post hoc analyses showed that the CA depleted group showed significantly longer escape latencies than DA and NE depleted animals as well as than controls throughout the test session, $p<0.05$.

Figure 6 depicts the percentage of trials during which animals in each treatment group failed to avoid the shock. The control animals rarely failed to avoid shock unlike animals in all treatment groups. An overall ANOVA revealed an effect of treatment, $F(3,45)=19.02$, $p<0.001$ and post hoc t tests revealed that control animals were significantly different from all other groups, $p<0.05$. Although CA depleted animals appeared to fail to avoid shock more than other treatment groups, these differences were not statistically significant.

DISCUSSION

The results of the present experiment add to the growing body of evidence which shows that neonatal CA depletion produces hyperactivity during ontogeny [9,15]. This report also agrees with others which suggest that the hyperactivity induced by neonatal 6-OHDA injections is a function of DA and not NE depletion [7, 13, 25]. In the present study, DA and CA depleted animals exhibited similar levels of hyperactivity which did not decline throughout the test session. In contrast NE depleted animals exhibited activity levels which were similar to control values, and decreased over time. Figure 2 of this report shows that differences in activity levels between treatment groups just begins to emerge in the second trial which is after 15 minutes, thus emphasizing the importance of using a test period which is longer than those utilized in other reports [12, 14, 16].

Although hyperactivity was seen in DA and CA depleted animals and not in NE depleted or controls, all 6-OHDA treated animals exhibited learning deficits. Such impairments in learning have previously been reported by many investigators for neonatally DA [15, 25, 28], CA [15] and NE [12, 14, 23] depleted animals. As mentioned (see Introduction), the absence of deficits in performance of NE depleted animals in an earlier experiment [16] may be explained by the relatively lenient criterion employed in this particular learning paradigm.

The data from the present experiment are particularly intriguing in that they show that the learning deficits observed in NE depleted animals are independent of changes in activity. Given that both T maze and Shuttlebox paradigms require an active escape response, deficits in this response could be a function of motor dysfunction rather than learning per se. The finding, however that NE depleted animals exhibited activity levels which were similar to controls and yet showed learning impairments suggests a role for NE in the ontogeny of avoidance learning.

The underlying explanation for why neonatally NE de-

pleted rats showed learning deficits has yet to be determined. Perhaps these animals are unable to selectively attend to the stimuli necessary for such learned associations. In an elaborate set of experiments, Mason *et al.* [11] have proposed that lesions of the dorsal NE bundle, which leads to significant reductions in cortical NE, produces animals with severe attentional deficits. While such an argument is compelling, a recent investigation reportedly failed to replicate this work [27]. Another possible explanation for deficits in avoidance learning displayed by a NE depleted rat is that they fail to avoid shock because they are less anxious [10]. Many investigations [8,18] have shown that lesions of the locus coeruleus, the nucleus containing NE cell bodies which innervate the cortex, leads to a decrease in fearfulness and anxiety in adult rats. Thus, neonatally NE depleted pups may have shown impairments in shock avoidance because they were less fearful of the noxious stimulus. Whether the learning deficits produced by early destruction of NE is a function of an attentional deficit, a decrease in anxiety, or an alteration in memory function must await future research.

In the present experiment early destruction of DA, NE as well as both CAs was performed and behavioral changes were observed. The purpose of the present experiment was to utilize identical behavioral measures for all treatment groups so that the effects of differential lesions could be directly compared. While this study succeeded in that regard it must be pointed out that the extent of neurotransmitter depletion was not identical for all groups. For example, although the NE depletion protocol was in fact selective for NE, cortical levels were 55% less than control values whereas the DA depletion protocol produced a 96% reduction in striatal DA. Thus, the comparisons in this study were necessarily made between moderate, albeit significant, NE depletion and severe (and also significant) DA depletion. Definitive conclusions regarding the differential effects of early DA and NE must await a study in which both the behavioral measures and the extent of neurotransmitter depletion is identical in both groups.

In conclusion, the present research may have some implications for clinical investigations, although one must always be cautious in extrapolating from research with laboratory animals to behavior in humans. Many researchers have sought to identify the neural correlates of the Attentional Deficit Disorder (ADD) of childhood, which is characterized by learning disabilities, short attention span and in many cases, hyperactivity [1]. These clinical investigations have yielded inconsistent findings; some studies show that ADD children have deficits in dopaminergic function [6,19], others report a noradrenergic deficit [26] and still another reports no deficits in the enzyme necessary for normal noradrenergic function [17]. These findings may appear inconsistent because the diagnosis of ADD in children is so broad that it includes a myriad of symptoms, each of which may be mediated by different neurotransmitter systems. Perhaps future clinical research on ADD can focus more on correlating specific behavioral symptoms, rather than a broad diagnosis, with neurotransmitter function. This possibility must be considered in light of the present finding which showed that neonatal lesions of the DA and NE systems in rats produced a constellation of behaviors which were distinctly unique.

REFERENCES

1. American Psychiatric Association. *Diagnostic and Statistical Manual of Mental Disorders,* (Third edition). Library of Contress Catalog Number 79-055868, 1980.
2. Anderson, G. M. and J. G. Young. Minireview: Applications of liquid chromatographic-fluorometric systems in neurochemistry. *Life Sci* **28:** 507–517, 1981.
3. Breese, G. R. and T. D. Traylor. Developmental characteristics of brain catecholamines and tyrosine hydroxylase in the rat: Effects of 6-OHDA. *Br J Pharmacol* **44:** 210–222, 1972.
4. Campbell, B. A., L. D. Lytle and H. C. Fibiger. Ontogeny of adrenergic arousal and cholinergic inhibitory mechanisms in the rat. *Science* **166:** 637–638, 1969.
5. Campbell, B. A. and P. D. Mabry. The role of catecholamines in behavioral arousal during ontogenesis. *Psychopharmacologia* **31:** 253–264, 1973.
6. Cohen, D. J., B. A. Shaywitz, J. G. Young and M. B. Bowers, Jr. Cerebrospinal fluid monoamine metabolites in neuropsychiatric disorders of childhood. In: *Neurobiology of Cerebrospinal Fluid,* edited by J. Wood. New York: Plenum Press, 1980.
7. Erinoff, L., R. C. MacPhail, A. Heller and L. S. Seiden. Age dependent effects of 6-hydroxydopamine on locomotor activity in the rat. *Brain Res* **164:** 195–205, 1979.
8. File, S. E., J. F. W. Deakin, A. Longden and T. J. Crow. An investigation of the role of the locus coeruleus in anxiety and agonistic behavior. *Brain Res* **169:** 411–420, 1979.
9. Fobes, J. L. and M. E. Olds. Effects of neonatal 6-hydroxydopamine treatment on catecholamine levels and behavior during development and adulthood. *Psychopharmacology* **73:** 27–30, 1981.
10. Gray, J. A., N. Davis, J. Feldon, J. N. P. Rawlens and S. R. Owen. Animals models of anxiety. *Prog Neurol Psychopharmacol* **5:** 143–157, 1981.
11. Mason, S. T. and S. D. Iversen. Theories of the dorsal bundle extinction effect. *Brain Res Rev* **1:** 107–137, 1979.
12. McLean, J. H., R. M. Kostrzewa and J. G. May. Behavioral and biochemical effects of neonatal treatment of rats with 6-hydroxydopa. *Pharmacol Biochem Behav* **4:** 601–607, 1976.
13. Miller, F. E., T. G. Heffner, C. Kotake and L. S. Seiden. Magnitude and duration of hyperactivity following neonatal 6-hydroxydopamine is related to the extent of brain dopamine depletion. *Brain Res* **229:** 123–132, 1981.
14. Morgan, D. N., J. H. McLean and R. M. Kostrzewa. Effects of 6-hydroxydopamine and 6-hydroxydopa on development of behavior. *Pharmacol Biochem Behav* **11:** 309–312, 1979.
15. Pappas, B. A., J. V. Gallivan, T. Dugas, M. Saari and R. Ings. Intraventricular 6-hydroxydopamine in the newborn rat and locomotor responses to drugs in infancy: No support for the dopamine depletion model of minimal brain dysfunction. *Psychopharmacology (Berlin)* **70:** 41–46, 1980.
16. Pappas, B. A. and S. K. Sobrian. Neonatal sympathectomy by 6-hydroxydopamine in the rat: No effects on behavior but changes in endogenous brain norepinephrine. *Life Sci* **14:** 887–898, 1974.
17. Rapoport, J. L., P. O. Quinn and F. Lamprecht. Minor physical anomalies and plasma dopamine beta hydroxylase activity in hyperactive boys. *Am J Psychiatry* **131:** 386–392, 1974.
18. Redmond, D. E. and Y. H. Huang. New evidence for a locus coeruleus-norepinephrine connection with anxiety. *Life Sci* **25:** 2149–2162, 1979.
19. Shaywitz, B. A., D. J. Cohen and M. B. Bowers, Jr. CSF monoamine metabolites in children with minimal brain dysfunction—Evidence for alteration of brain dopamine. *J Pediatr* **90:** 67–71, 1977.

20. Shaywitz, B. A., J. W. Gordon, J. H. Klopper and D. A. Zelterman. The effect of 6-hydroxydopamine on habituation of activity in the developing rat pup. *Pharmacol Biochem Behav* **6:** 391–396, 1977.
21. Shaywitz, B. A., J. H. Klopper and J. W. Gordon. Methylphenidate in 6-hydroxydopamine treated developing rat pups. *Arch Neurol* **35:** 463–469, 1978.
22. Shaywitz, B. A., J. H. Klopper, R. D. Yager and J. W. Gordon. Paradoxical response to amphetamine in developing rats treated with 6-hydroxydopamine. *Nature* **261:** 153–155, 1976.
23. Shaywitz, B. A., C. S. Kretschmar, P. Levitt, M. H. Teicher, G. M. Anderson, J. G. Young and D. J. Cohen. Selective damage to noradrenergic systems in the developing rat pup affects escape and avoidance performance but does not influence activity. Submitted for publication, 1983.
24. Shaywitz, B. A., M. H. Teicher, D. J. Cohen, G. M. Anderson, J. G. Young and P. Levitt. Dopaminergic but not noradrenergic mediation of hyperactivity and performance deficits in the developing rat pup. *Psychopharmacology (Berlin)*, in press.
25. 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.
26. Shekim, W. O., H. Dekirmenjian, J. L. Chapel, J. Javaid and J. M. Davis. Norepinephrine metabolism and clinical response to dextroamphetamine in hyperactive boys. *J Pediatr* **95:** 389–394, 1979.
27. Tombaugh, T. N., B. A. Pappas, D. C. S. Roberts, G. J. Vichers and C. Szostak. Failure to replicate the dorsal bundle extinction effect: Telencephalic norepinephrine depletion does not reliably increase resistance to extinction but does augment gustatory neophobia. *Brain Res* **261:** 231–242, 1983.
28. Weldon, D. A., R. S. Wool, M. H. Teicher, B. A. Shaywitz, D. J. Cohen and G. M. Anderson. Effects of Apomorphine on appetitive conditioning in 6-hydroxydopamine treated rat pups. *Pharmacol Biochem Behav* **17:** 1281–1284, 1982.