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BRIEF COMMUNICATION

No Increase in Reaction-Time After Lesion of the Dorsal Noradrenergic Bundle

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BRENNER, E., B. C. THEUNISSE, M. MIRMIRAN AND J. VAN DER GUGTEN. *No increase in reaction-time after lesion of the dorsal noradrenergic bundle.* PHYSIOL BEHAV 39(5) 653–656, 1987.—In this study we examined whether lesions of rats' dorsal noradrenergic bundles affect their reaction times to temporally unpredictable stimuli. Injection of 6-hydroxydopamine into rats' dorsal bundles drastically reduced the noradrenaline content of their cerebral cortex. Nevertheless, 6OHDA treated rats could still react as quickly as controls. Moreover, the treatment did not affect the efficiency with which rats performed the task, even when they were forced to respond very quickly.

Noradrenaline Reaction-time Attention Dorsal bundle 6-Hydroxydopamine Rat

THE dorsal noradrenergic fibre bundle has been attributed a role in attention and arousal (e.g., [1,7]). However, behavioural studies designed to elucidate its role in attention have produced conflicting results. Rats with dorsal bundle lesions were found to be more distractable than controls when performing a simple maze task [13] but not when drinking [5,6]. Impaired reversal shift and enhanced non-reversal shift after dorsal bundle lesions were interpreted as indicating less exclusive attention to the relevant stimuli [9,10]. However, these effects were not found in other similar studies [11]. Such lesions have also been reported to increase both latent learning [10] and resistance to extinction [8], but do not appear to do so consistently ([2,17] and [16], respectively).

A task requiring a reaction to spatially unpredictable stimuli showed that rats with dorsal bundle lesions were less efficient than controls when the stimuli were temporally unpredictable as well: presenting unpredictable stimuli at a high rate decreased the efficiency of the rats' performance [4]. In studies with human subjects, attention to stimuli has been quantified by measuring reaction-times ([3] (chapter 7), [12]), as well as by determining response efficiency (number of omissions and of responses when no stimulus is presented; see [3]). We reasoned that determining the thresholds of rats' reaction times may reveal differences in their attention to temporally unpredictable stimuli. As even modest depletion of dopamine has been shown to reduce the speed with which rats are able to perform a motor response [15], a second task was used to make sure that the speed with which they could perform the required motor response was not affected.

METHOD

Thirty-one male Wistar rats were deprived of food for all but one hour per day immediately after behavioural testing. Tests were carried out at the beginning of the light period in four Grason-Stadler (model 1111-L) conditioning chambers, with a lever on either side of a pellet retrieval unit, and a lamp providing bright light above and slightly to one side of each lever. Each chamber was inside a (model 1101) research chest with a fan providing fresh air. Grason-Stadler programming equipment (1200 series) was used for controlling the experimental procedures and collecting the data. In the first task the rats obtained food pellets by pressing the left lever (which made the lamp above it go OFF) and then responding on the right lever when the right lamp went ON. There was an unpredictable interval, that varied at random between 3 and 6 seconds, between the moment the left lever

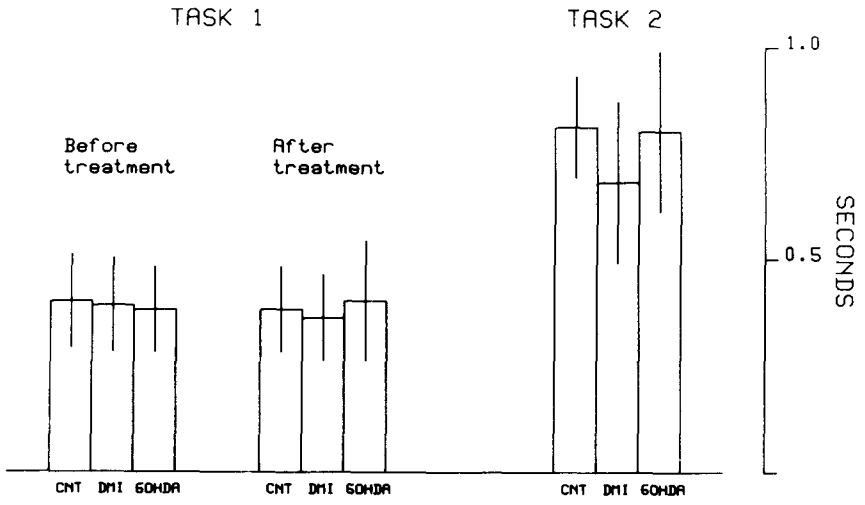

FIG. 1. Mean thresholds (with standard deviations) for the 8 saline treated controls (CNT), the 7 desmethylimipramine pretreated rats (DMI) and the 15 rats that received 6-hydroxydopamine without pretreatment (6OHDA). The rats performed task 1 both before and after treatment, whereas task 2 was only performed after treatment.

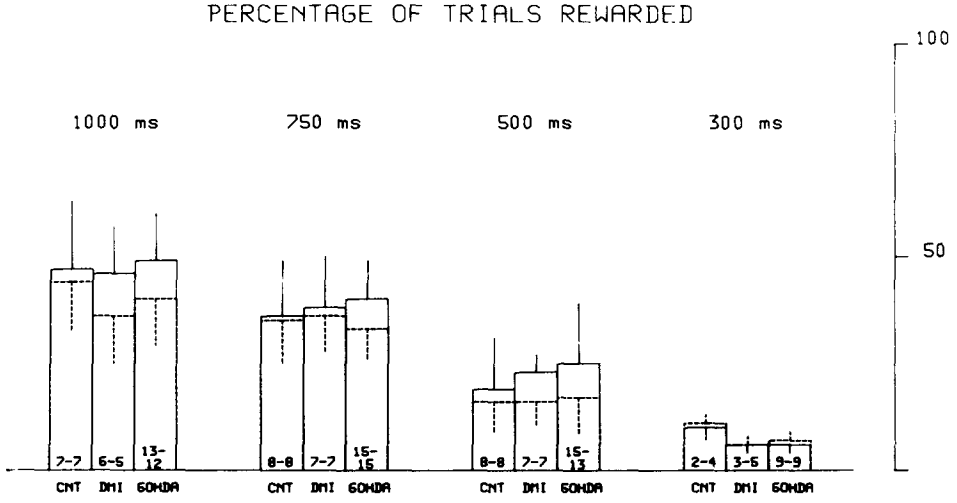

FIG. 2. Mean percentage of presses on the left lever that resulted in the rats' obtaining the reward. Continuous lines: last ten days of task 1 after treatment. Dashed lines: last ten days on the same task before treatment. Results are split into sessions in which the rats had to respond within 1000, 750, 500 and 300 msec after stimulus onset. As the time within which the rats had to respond depended on their performance, not all rats were tested at all levels. The number of rats that provided data at each level (before-after treatment) is shown within the bars. The means and standard deviations were calculated after one value was determined for each rat's performance at the specified level during the appropriate period.

was pressed and illumination of the lamp on the right. When the right lamp was ON, the rats were rewarded if they pressed the right lever. However, the right lamp went OFF again after a certain time period. Sequences ended either by the rat pressing the right lever—even if this was before the right lamp went ON—or by the right lamp going OFF before the rat pressed the lever. Between sequences the lamp on the left was ON. The time allowed for responding was reduced using a staircase procedure until each rat's threshold was found: if the rat pressed the right lever before the right lamp went OFF on more occasions than those on which it was too late (at least 30 rewards), the time that the right lamp remained ON was decreased from 100 to 75, 50, 30, 20, 15, 10, 7.5, 5, 3, 2, 1, 0.75, 0.5 and 0.3 seconds. Otherwise, the time was increased by two such steps. Rats were left in the box until they either obtained 60 pellets or allowed the left lamp to be ON for 15 minutes. The lowest duration of right lamp illumination that the rat reached was considered its threshold. Furthermore, for performance at several response requirements, the number of sequences on which the rat obtained the reward was expressed as a percentage of the total number of sequences that were initiated.

TABLE 1

MEAN NORADRENALINE AND DOPAMINE CONTENTS IN ng/mg WET WEIGHT (WITH STANDARD DEVIATIONS)

	Saline	DMI+6OHDA	6OHDA
Number of Rats	8	7	15
Noradrenaline			
Cerebral Cortex	0.65 (0.19)	0.46 (0.27)	0.08 (0.05)
Cerebellum	0.57 (0.13)	0.60 (0.11)	0.67 (0.18)
Spinal Cord	0.39 (0.05)	0.40 (0.06)	0.40 (0.07)
Dopamine			
Cerebral Cortex	0.26 (0.07)	0.24 (0.08)	0.29 (0.13)
Spinal Cord	0.06 (0.01)	0.06 (0.01)	0.06 (0.01)

After 50 days of training on the first task, the rats were randomly divided into 3 groups. Four micrograms of the neurotoxin 6-hydroxydopamine HCl (6OHDA), dissolved in 2 microliters of saline containing 0.1 mg/ml ascorbic acid, was injected during 4 minutes into 15 rats' dorsal bundles under fentanyl anaesthesia ("Hypnorm," Duphar B.V.), and the injection needle (Hamilton, 0.7 mm diameter) was left in place for 2 more minutes. The coordinates for the injections were: 6 mm posterior to bregma, 1 mm to each side of the midline and 5 mm below the cortical surface. Eight control rats received the same injections without the 6OHDA. A second group of 8 control rats were injected with 6OHDA, but their noradrenergic fibres were protected by a noradrenergic uptake blocker, desmethylimipramine (DMI; 20 mg/kg half an hour before injecting 6OHDA). One of these DMI pretreated rats died within 24 hours after treatment. The rats' thresholds were re-determined on the same task during 25 more days (starting with the right lamp remaining illuminated for 5 seconds, ten days after the injections). Once the rats completed the first task—after treatment—they were tested on the second task, in which there was no interval between the moment they pressed the left lever and illumination of the right lamp. The same staircase procedure was used to determine how quickly the rats could press the right lever after pressing the left one. After the behavioural tests, rats were decapitated and their cerebral cortex, cerebellum and spinal cord were immediately dissected, frozen and stored −80°C. Noradrenaline and dopamine contents were determined radioenzymatically [18].

RESULTS

The mean thresholds on both tasks are shown in Fig. 1. All rats had reached a threshold of either 500 or 300 msec on the first task before the injections took place. The rats' thresholds as well as the number of times that they reached this threshold were used to rank them for comparison of the 3 groups with the Kruskall-Wallis H-test. The rat that most often reached the shortest duration of right lamp illumination (300 msec in task 1) was assigned the rank "1." All rats that reached this duration were then ranked according to how often they had done so, before continuing with successively shorter durations of right lamp illumination. The treatment did not result in a significant difference between the 3 groups of rats' performance, either on the first, $H(2)=1.04$, or on the second, $H(2)=2.32$, task. Furthermore, for task 1, a high correlation between the rats' ranks before and after treatment (a Spearman rank correlation coefficient of 0.83; calculated disregarding grouping) confirms that the rats' relative levels of performance were not affected by the fact that they had received different treatments. All but 2 of the 6OHDA treated rats and all of the controls regained their pre-operative thresholds within 6 weeks.

In Fig. 2, the percentage of trials that were rewarded when rats were required to respond within 1000, 750, 500 and 300 msec are shown for the 16th to the 25th session of task 1 after treatment, and for the last 10 sessions before treatment. Rats were evaluated on this measure of response efficiency with t-tests. 6OHDA-treated rats were significantly less efficient than saline treated controls when forced to respond within 300 msec, $t(11)=2.60$, $p<0.05$, but their performance did not differ from that of the DMI pretreated controls. Furthermore, during the last 10 days before treatment, the 2 saline-treated rats that had reached the 300 msec level already performed more efficiently than their 6OHDA treated counterparts, $t(9)=2.31$, $p<0.05$. At 500, 750 and 1000 msec there were no significant differences between the groups. In no group, and under none of the required response speeds, did rats perform significantly less well after treatment than before. The 6OHDA treated rats even performed more efficiently at 750 msec and the DMI pretreated controls did so at 500 msec. This may be due to additional experience with the task.

The lesion reduced the noradrenaline content of the cerebral cortex to 12% of that of the saline injected controls, $t(21)=10.44$, $p<0.001$, and to 17% of that of the DMI treated rats, $t(20)=5.02$, $p<0.001$ (Table 1). The values for the DMI treated controls did not differ significantly from those of the saline treated controls, $t(13)=1.48$, $0.1<p<0.2$. Neither the noradrenaline contents of cerebellum and spinal cord nor the dopamine contents of cerebral cortex and spinal cord were affected. The cerebellum contained too little dopamine for it to be measured reliably.

DISCUSSION

The 6OHDA treatment appears to have successfully and selectively destroyed the dorsal noradrenergic bundles. Furthermore, pretreatment with DMI protected the fibres from the toxin reasonably effectively, providing us with an additional control group that can be expected to exhibit any 6OHDA-induced impairments that are not the result of destruction of noradrenergic fibres. After treatment, the rats were tested as soon as possible in order to limit the time available for functional compensation for the destroyed innervation. The behavioural results show that the speed with which rats can respond to a temporally unpredictable bright visual stimulus is not affected by a lesion that reduces the cortical noradrenaline content to 12% of that of the controls. The only significant impairment in the 6OHDA treated rats' behaviour was a lower efficiency at the fastest required response level. However, as the DMI group's performance resembled that of the 6OHDA treated group, rather than that of the saline treated group, and as this difference was already present before treatment, we can be reasonably certain that the difference is not due to the destruction of noradrenergic fibres.

Noradrenaline has been reported to increase the signal to noise ratio within the brain (see [14]). A decrease in the signal to noise ratio after destruction of the noradrenergic

innervation may increase the time needed to make a distinction between the presence and absence of a stimulus, leading to slower reactions or less efficient performance when a quick decision is required. Lesioned rats would have to adopt a more risky response criterion when forced to react quickly [3], and therefore to press more often in the absence of a stimulus. This was not the case. Destroying the noradrenergic innervation of rats' forebrains neither made them miss more stimuli, which would have affected their thresholds, nor did it cause them to press the right lever more often in the absence of a stimulus, i.e., to perform less efficiently.

It may be concluded, that an intact dorsal noradrenergic fibre bundle is not essential for the attentive requirements of the present task. It may, however, prove to be necessary when using less conspicuous or less predictable stimuli. Furthermore, we cannot be certain that even more extensive lesions also would not have impaired performance on this task. Similarly, testing the rats sooner after the lesions, in order to limit the time available for compensatory processes, may also have revealed behavioural deficits. However, the present lesion was so conspicuously ineffective at modifying the rats' performance, that we do not expect that slight changes in depletion or timing would be enough to change the results.

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