EFFECTS OF PERINATAL HYPOTHYROIDISM ON SHIFT ATTENTION IN RATS

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Abstract

Pregnant rats were treated with the anti-thyroid drug methimazole [0.02% (w/v) in drinking water] from gestational day 15 to postnatal day 21. The offspring received behavioral testing using a shift attention task. The testing started with the presentation of one of the two targets. After 5 s, a lever was inserted to the same side as the presented target (ipsilateral trials). Rats learned to pay attention to the target-presented side and respond on the inserted lever. The contralateral trials were then introduced. Since the contralateral trials inserted a lever to the opposite side to the presented target, rats were required to turn their attention from the target-presented side to the opposite side to respond. The ipsilateral and contralateral trials were presented pseudo-randomly with probabilities of 80% and 20%, respectively. On the ipsilateral trials, both the treated and untreated rats were able to respond correctly on the lever more than 90% of the time with quick and specific reaction times. On the contralateral trials, however, treated rats did not respond on the contralaterally inserted lever or they responded sporadically with a wide range of reaction times. This indicates that the rats did not shift their attention toward the opposite side within a fixed interval. The untreated rats responded correctly with shorter reaction times compared to the treated rats and were able to shift their attention toward the opposite side within a fixed interval. Taken together, we conclude that perinatal hypothyroidism affects shift attention in rats.

Introduction

Polychlorinated biphenyls (PCBs) are persistent organic pollutants that affect thyroid hormone systems.[7] Thyroid hormone is essential for normal brain development because it regulates neuronal proliferation, migration, and differentiation.[8] Reports have shown that the children of mothers who ingested fish from Lake Michigan after contamination with PCBs have attention deficit behavior.[2, 3] Thus, it is possible that PCB-induced hypothyroidism might cause attention deficit behavior in children.

The relationship between hypothyroidism and attention deficit behavior has been experimentally studied in animal model systems. For instance, Holene et al.[1] treated rats with PCBs and showed that they unnecessarily visited a food tray, a sign of inattention. Similarly, after treating with propylthiouracil, an anti-thyroid drug, Negishi et al.[6] trained rats in a shock avoidance task and demonstrated increased movement across the partition between two avoidance compartments, also a sign of inattention. In addition, Wada used a target detection task to study attention deficit behavior and demonstrated that perinatal hypothyroid rats do not react quickly to a presented target.[9]

Attention ability consists of several sub-functions: focused attention, sustained attention, shift attention, and divided attention. Improving upon the target detection task, Wada introduced a retractable lever to a skinner box to study focused and sustained attention.[10] He observed that hypothyroid rats did not quickly focus attention or sustain attention on the target. Thus, the aim of this study was to determine whether hypothyroid rats are able to turn attention from one target side toward the other target side (i.e., shift attention).

Materials and methods

Twenty pregnant Wistar rats were purchased on gestational day 8. Rats were housed in individual cages under a 12-h light/dark cycle (light, 19:00–07:00 h; dark, 07:00–19:00 h). Room temperature was maintained at 22 ± 2°C, and relative humidity was 50 ± 10%. The rats were randomly assigned to a treated (n=10) or untreated (control) group (n=10). After dissolving in distilled water, the anti-thyroid drug methimazole [0.02% (w/v)] was administered to the treated rats via drinking water beginning on gestational day 15 until postnatal day 21. Offspring from the untreated rats were weaned at postnatal day 21; however, offspring from the treated rats were weaned at postnatal day 28 due to developmental delay. After weaning, one male offspring was sampled from each dam; thus, 10 male offspring were assigned to the treated and untreated groups. These rats were individually housed and received food and water *ad libitum* until eight weeks of age. All rats were then placed

under restricted food conditions and maintained at 85% of their free-feeding body weights. Following behavioral testing for focused and sustained attention, shift attention testing began at seven months of age. All experiments were conducted during the dark period. We note that one treated rat died before the testing.

Five standard operant chambers with two retractable levers were used. The target was a light emitting diode mounted above each retractable lever. A speaker was placed outside of the chamber, and white noise (70dB) was presented to mask external sound. The chamber was set in an isolation box designed to attenuate external light and sound. Experiments and data recording were controlled by a personal computer.

The behavioral testing for shift attention consisted of ipsilateral and contralateral trials. The ipsilateral trial started with the presentation of one of the two targets. After 5 s, the lever on the same side as the presented target was inserted for 3 s. A correct response on the lever resulted in a reward of a 50 mg-food pellet. The target was then removed and the lever was retracted. The next trial started after a 30 s inter-trial interval (ITI). If no response occurred on the lever within 3 s, the target was removed and the lever was retracted. Again the next trial started after a 30 s ITI. After the rats learned to respond on the ipsilaterally inserted lever, the contralateral trials were introduced and followed the same pattern as the ipsilateral trials. The contralateral trial started with the presentation of one of the two targets. After 5 s, the lever on the opposite side to the presented target was inserted for 10 s. A correct response to the lever resulted in a reward of a 50 mg-food pellet. The target was then removed and the lever was retracted. The next trial started after a 30 s ITI. If no response occurred on the lever within 10 s, the target was removed and the lever was retracted. The next trial started after another 30 s ITI. The ipsilateral and contralateral trials were presented pseudo-randomly with probabilities of 80% and 20%, respectively. The behavioral testing for shift attention consisted of 40 ipsilateral and 10 contralateral trials each day and continued for nine days.

This research was carried out with the approval of Hokkaido University. The environmental conditions complied with The Guide for the Care and Use of Laboratory Animals (Hokkaido University).

Results and discussion

On the ipsilateral trials, both the treated and untreated rats responded correctly on more than 90% of the trials from training day 1 through 9. Although the percentage for the treated rats was slightly lower than those for the untreated rats [$F(1,17)=9.013$, $p<0.01$], multiple comparison tests did not reveal significant differences between the treated and untreated rats for each training day. On the contralateral trials, however, the treated rats did not exhibit increased percentages of correct responses compared to the untreated rats (Fig. 1). There was a significant difference between the treated and untreated rats [$F(1,17)=8.568$, $p<0.01$], and the treated rats exhibited significantly lower percentages of correct responses compared to the untreated rats on training day 2 to 7 using multiple comparison tests.

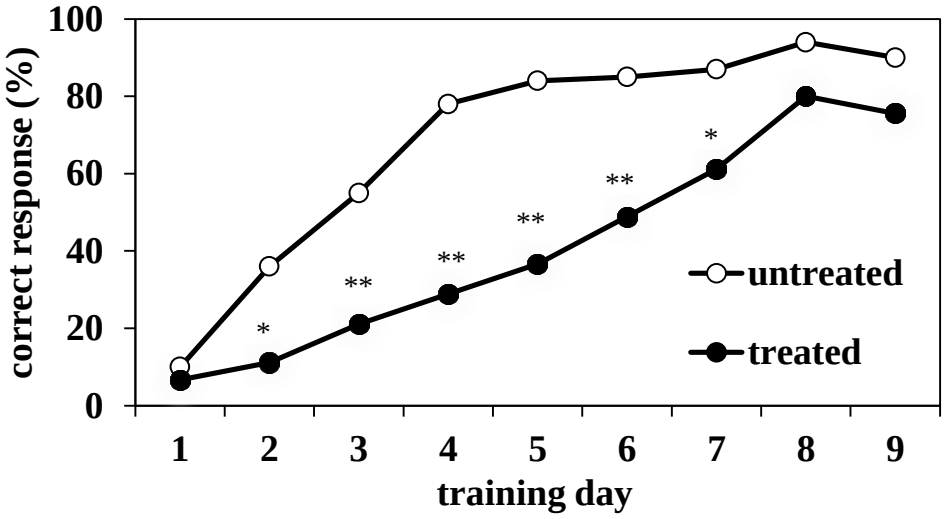

Figure 1. Mean percentage of correct responses in the contralateral trials

** p<0.01 and * p<0.05 compared to the untreated rats

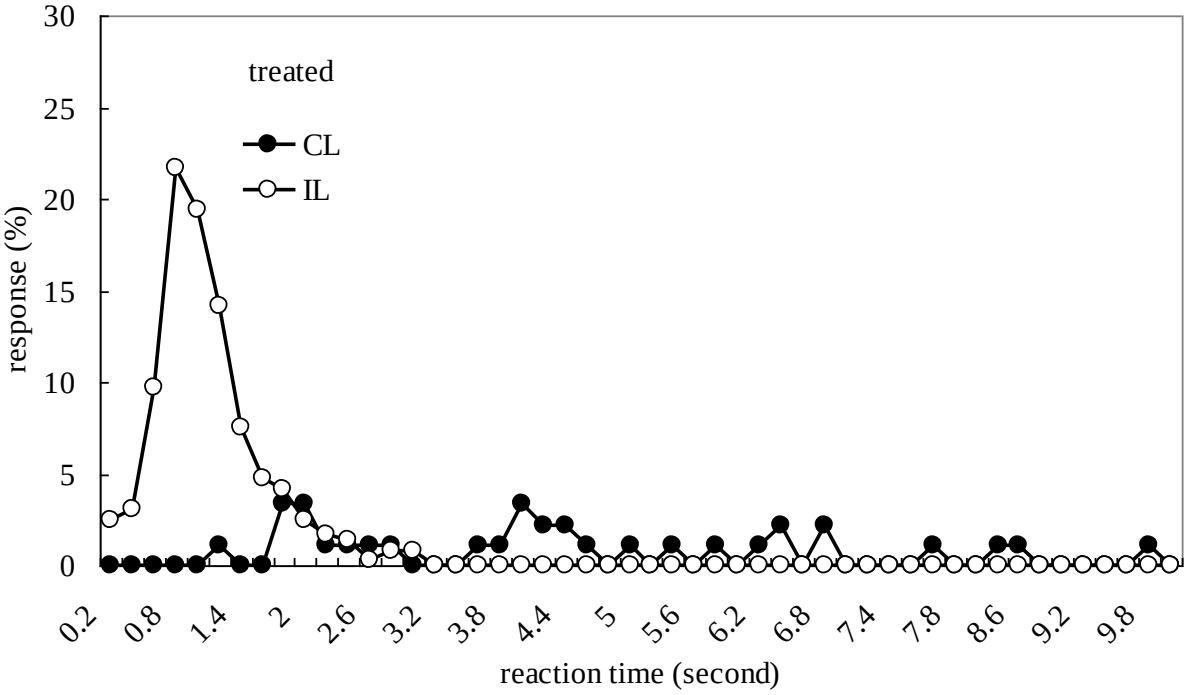

Figure 2. Reaction time distribution for the treated rats on training day 5
The horizontal axis represents reaction time with a segment of 0.2 s.
CL, contralateral; IL, ipsilateral

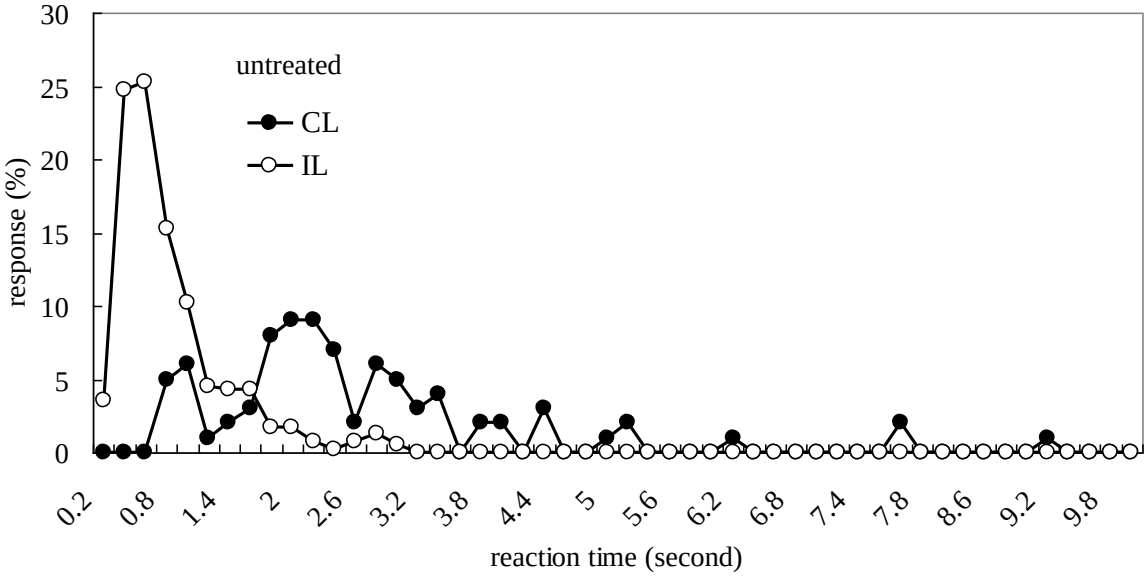

Figure 3. Reaction time distributions for the untreated rats on training day 5
The horizontal axis represents reaction time with a segment of 0.2 s.
CL, contralateral; IL, ipsilateral

Reaction time distributions on training day 5 are displayed in Fig. 2 (treated rats) and Fig. 3 (untreated rats). When the lever was inserted to the same side as the presented target (ipsilateral trial), the treated and untreated rats frequently responded with a reaction time of 0.6 s to 1.2 s and 0.2 s to 0.8 s, respectively. The means and SDs of the reaction times were 1.01±0.51 s and 0.73±0.53 s for the treated and untreated rats, respectively. We observed that when rats approached the target-presented side, they clung to the side until lever insertion and then quickly responded on the lever. This observation indicates that both the treated and untreated rats were able to selectively pay attention to the target-presented side and quickly respond on the lever.

For the contralateral trials, rats approached the target-presented side and clung to the side. After the lever was inserted on the opposite side, the rats moved from the target-presented side to the opposite side to respond. The rats had to shift their attention away from the target-presented side and toward the opposite side where the lever was inserted. The untreated rats exhibited increased percentages of correct responses for the contralateral trials, which resulted in greater than 80% correct responses after training day 5 (Fig. 1). Moreover, the reaction time distribution displayed a clear peak between 0.6 s to 3.4 s (Fig. 3). The mean and SD of the reaction time was 2.56±1.54 s for the untreated rats. This suggests that the untreated rats were able to abandon their attention toward the target-presented side within a fixed interval and turn their attention toward the opposite side. In contrast, the treated rats had lower percentages of correct responses until training day 7 and only reached 80% on training day 8 (Fig. 1). The rats did not respond on the contralaterally inserted lever or they responded sporadically with a wide range of reaction times (Fig. 2). The mean and SD of the reaction time was 4.35±2.24 s for the treated rats. We found a slower reaction time and larger SD for the treated rats compared to the untreated rats. The same pattern of reaction time distribution was found until training day 9.

Since hypothyroid rats do not alternate their responses between the two levers and persist on the same lever,[3, 4] the treated rats in our study might not be able to abandon attention after they have fixed their attention on the target-presented side. However, we observed that hypothyroid rats leave the target-presented side and approach a food tray or the other target side during a sustained attention task.[10] Thus treated rats might turn their attention toward inappropriate stimuli. Taken together, we conclude that treated rats had difficulties shifting attention from one target toward another appropriate target. Further studies are necessary to clarify effects of perinatal hypothyroidism on shift attention.

Acknowledgements
The authors thank Mss. S. Shigemi, M. Shimode, and N. Taniguchi for their technical assistance. This study was supported by the Long-range Research Initiative of the Japan Chemical Industry Association.

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