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Research report

Effects of exposure to 50 Hz magnetic field of 1 mT on the performance of detour learning task by chicks

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Abstract

In the present study, we examined the effects of exposure to an extremely low-frequency magnetic field of 1 mT intensity on learning and memory in Lohmann brown domestic chicks using detour learning task. These results show that 20 h/day exposure to a low-frequency magnetic field induces a significant impairment in detour learning but 50 min/day exposure has no effect.

Keywords: Lohmann brown domestic chick; Detour learning; Low-frequency magnetic field; Learning and memory

1. Introduction

The exposures to extremely low-frequency magnetic fields (ELFMF) in our environment have dramatically increased, which includes both occupational exposure and general exposure to sources, such as power lines, household electrical wiring and medical devices. This produced a social alarm on the possible adverse effects of magnetic fields on human health and stimulated a number of investigations on the biological effects of ELFMF on living organisms [13]. Some reports show that ELFMF may interfere on the activity of the brain [5,7,12], generate behavioral and cognitive disturbances [8,15,29], increase the risk of neurodegenerative diseases in humans [25] and produce deficits in attention, perception and spatial learning in rats [16]. Furthermore, the developing central nervous system (CNS) exhibits even higher sensitivity to ELFMF [4]. Prenatal or perinatal exposure to ELFMF decreases the density of neurons in the medial preoptic nucleus, affects some sexually dimorphic structures, and impairs scent marking and inter-male fighting behaviors during adulthood [24,27].

Detour behaviour is the ability of an animal to reach a stimulus (goal) when there is an obstacle between the subject and the stimulus. Detour learning in chicks has been used as a functional test for the development of nervous system [29,30]. Since ELFMF may interfere with spatial learning, and detour learning task in chicks is an excellent model to study the development of spatial memory, the present study used detour learning task in Lohmann brown domestic chicks to investigate the effects of ELFMF on the spatial learning and development of CNS. Since the sensitive phase for the development of spatial memory is around post hatch day 11 [6], the domestic chicks were exposed to 50 Hz magnetic fields at 1 mT for 60 min or 20 h/day before and after post hatch day 11 and then detour learning task was performed and evaluated.

2. Materials and methods

2.1. Subjects and their treatment

32 Lohmann brown domestic chicks obtained from a commercial hatchery when they were only a few hours old were used. On arrival in the laboratory, domestic chicks were reared in rectangular cages (50 cm × 25 cm, with 25 cm high walls) at controlled temperature 30.8–35.8 °C, with food and water available ad libitum. The domestic chicks were exposed to a photoperiod of 12-h

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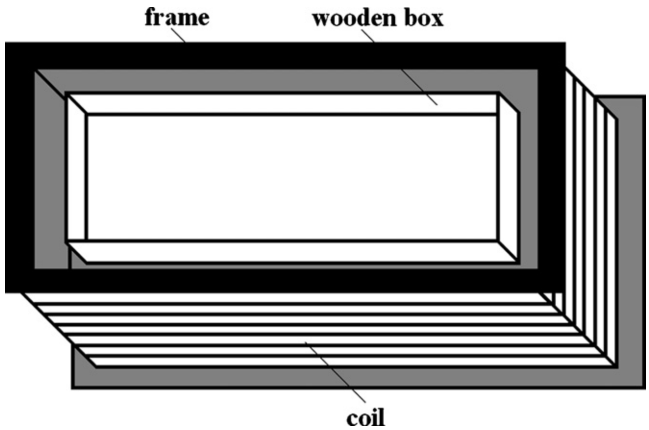

Fig. 1. Schematic diagram of electromagnetic field system and the exposure area. Note that the coils are wrapped horizontally around a plastic frame and the space inside the coils is the exposure area. During exposure, chicks were placed in a wooden box.

light and 12-h dark cycle. On post hatch day 7, domestic chicks were weighed, numbered and randomly assigned to four treatment groups ($n = 8$/group): (1) exposure to magnetic field 20 h/day after detour task learning; (2) exposure to sham magnetic field 20 h/day after detour task learning; (3) exposure to magnetic field 50 min/day after detour task learning; (4) exposure to sham magnetic field 50 min after detour task learning.

2.2. Magnetic field exposure system

As described earlier [17], the electromagnetic field was generated by a single coil of four layers, each having 250 turns. Each layer was wrapped horizontally above the previous layer around a 70 cm × 40 cm × 43 cm plastic frame. The coil was connected to a waveform generator for modulating the frequency and intensity of the electromagnetic field. By varying the input current to the coil, the flux density of electromagnetic fields in exposure area can be adjusted from the ambient level to the maximum coil-designed electromagnetic field strength of 14 mT.

The exposure area (60 cm × 30 cm × 43 cm) was inside the coil. During exposure, domestic chicks were placed in a wooden box (50 cm × 25 cm, with 25 cm high walls) which was mechanically isolated from the magnet and rested on a freestanding wood. The electromagnetic field system and the exposure area are shown in Fig. 1. The variation of the electromagnetic fields in the wooden box as determined by actual measurement was ±4.5% of the mean.

2.3. Detour learning

Domestic chicks were trained using the detour learning procedure described by Bollweg and Sparber [1]. The detour learning apparatus is a fluorescently illuminated wooden enclosure with a lid, separated into two compartments (social and isolation sides) by a Plexiglas wall. To return to the social side, isolated subjects must turn away from the transparent wall and detour through the open tunnel (Fig. 2). Under these conditions the opportunity for access to food and brood mates are appropriate stimuli for reinforcing the detour response, resulting in shorter latencies as subjects learn to detour. Four domestic chicks (one from each treatment group) were randomly selected from the community brooder and placed on the social side of the detour apparatus, which contained a plate with a small amount of moistened domestic chick food. Subjects were allowed access to the food and social reinforcement for 30 s, after which one was selected and placed in the center of the isolation side of the apparatus. This subject was allowed 180 s to face away from the reinforcing complex and detour through the open tunnel. If no detour response was made during this time, its latency was recorded as the maximum 180 s and the subject was gently guided through the tunnel with a wooden ruler, terminating the trial [1]. This sequence was repeated with the next subject until each of the four domestic chicks had received one trial. The colored, numbered leg bands allowed the experimenter to control for

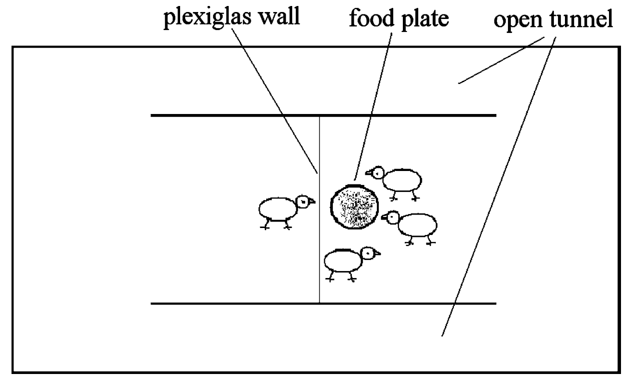

Fig. 2. Schematic diagram of detour learning apparatus. Note that the apparatus is separated into two compartments by a Plexiglas wall. To return to the social side, isolated subjects must turn away from the transparent wall and detour through the open tunnel.

order effects by systematically rotating the sequence of testing for each clutch of four domestic chicks. Detour learning task started on post hatch day 9 and is performed daily from post hatch day 9 to post hatch day 15. After 7 days of testing each animal received seven trials. Before each detour learning test, the animals were deprived of food for 12 h and weighted. After learning test, animals were placed in a wooden box (50 cm × 25 cm, with 25 cm high walls) with water and ad lib domestic chick food for about 2 h and then were exposed to 50 Hz magnetic field at 1 mT or sham magnetic field for 20 h or 50 min.

2.4. Statistical analysis

All data were analyzed using a statistical package for social sciences (SPSS10.0). The effects of ELFMF on spatial memory were determined by two-way repeated-measures ANOVA, followed by post hoc Newman-Keuls multiple comparison. Data were presented as mean ± S.E.M.

3. Results

Fig. 3 shows the detour learning results for exposure to sham or magnetic field 50 min/day. Compared to 50 min/day exposure to sham magnetic field, 50 min/day exposure to ELFMF had no effect on detour response latency for each trial. The average response latency for the first detour trial was around 170 ms. After two trials, the responses latency was significantly reduced

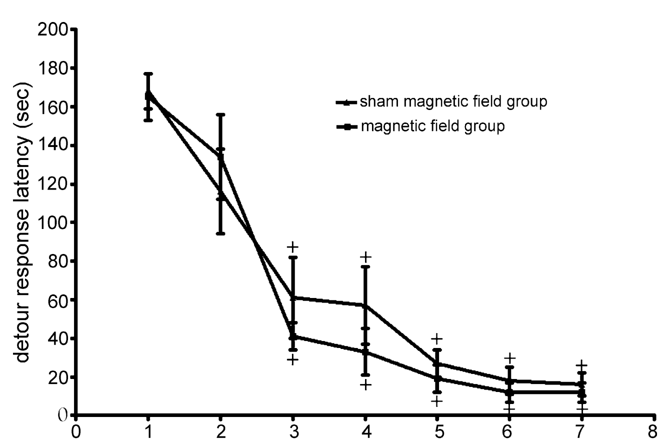

Fig. 3. Effects of exposure to low-frequency magnetic field 50 min/day on detour learning. Each point depicts the mean latency(s) for a group of eight chicks. (+) significant difference among trials, $p < 0.01$ or $p < 0.05$ (Newman-Keuls).

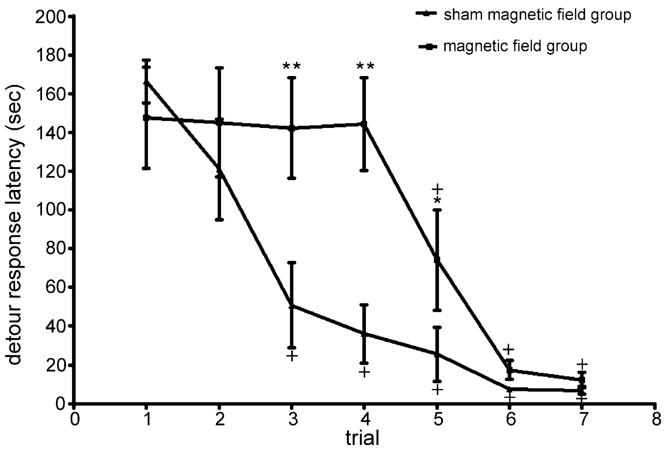

Fig. 4. Effects of exposure to low-frequency magnetic field 20 h/day on detour learning. Each point depicts the mean latency(s) for a group of eight chicks. (*) significant effect of ELFMF, $p<0.05$; (**) significant effect of ELFMF $p<0.01$; (+) significant difference among trials, $p<0.01$ (Newman-Keuls).

to 61 ms in ELFMF and 78 ms in sham exposure group. The response latencies of trials 3, 4, 5, 6 and 7 were significantly lower than that of trials 1 and 2 in both sham and ELFMF group (trial 1 versus trial 3; trial 1 versus trial 4; trial 1 versus trial 5; trial 1 versus trial 6 and trial 1 versus trial 7; $p<0.01$; trial 2 versus trial 3 and trial 2 versus trial 4; $p<0.05$; trial 2 versus trial 5; trial 2 versus trial 6 and trial 2 versus trial 7; $p<0.01$). There is no significant difference in response latency among the trials 3, 4, 5, 6 and 7 in both groups. The two-way repeated-measures ANOVA analysis showed that there was a main effect of trials ($F(6,111)=24.24$, $p<0.0001$), but there was no main effect of treatment ($F(1, 111)=0.12$, $p=0.72$) and no interaction between trial and treatment ($F(6,111)=0.26$, $p=0.95$).

Fig. 4 shows the detour learning results for exposure to sham or magnetic field 20 h/day. Compared to 20 h/day exposure to sham magnetic field, 20 h/day exposure to ELFMF significantly delayed the detour learning. In consistent with the 50 min/day exposure to sham or ELFMF, 20 h/day exposure to sham had no effect on detour learning. After two trials, the response latencies were significantly reduced in 20 h/day sham exposure group (trial 1 versus trial 3; trial 1 versus trial 4; trial 1 versus trial 5; trial 1 versus trial 6 and trial 1 versus trial 7; $p<0.01$; trial 2 versus trial 3 and trial 2 versus trial 4; trial 2 versus trial 5; trial 2 versus trial 6 and trial 2 versus trial 7; $p<0.01$). But in ELFMF exposure group, the response latencies were significantly reduced after four trials (trial 1 versus trial 5; trial 1 versus trial 6; trial 1 versus trial 7; trial 2 versus trial 5; trial 2 versus trial 6; trial 2 versus trial 7; trial 3 versus trial 5; trial 3 versus trial 6; trial 3 versus trial 7; trial 4 versus trial 5; trial 4 versus trial 6 and trial 4 versus trial 7; $p<0.01$). Furthermore, the response latencies of trials 3, 4 and 5 in ELFMF exposure group were significantly longer than that in sham exposure group (trial 3 and trial 4, $p<0.01$; trial 5, $p<0.05$). The two-way repeated-measures ANOVA analysis showed that there was a main effect of trials ($F(6,111)=52.93$, $p<0.0001$), a main effect of treatment ($F(1, 111)=7.70$, $p=0.01$) and a significant interaction between trial and treatment ($F(6,111)=9.05$, $p<0.0001$).

Table 1
Chick body weight after exposure to magnetic field or sham magnetic field

Treatment	Body weight (g)
Exposure to magnetic field 20 h/day	58.4 ± 2.0
Exposure to sham magnetic field 20 h/day	58.9 ± 1.8
Exposure to magnetic field 50 min/day	60.6 ± 2.0
Exposure to sham magnetic field 50 min	59.7 ± 2.2

Values represent the mean weight (g) ± S.D. of eight chicks per group after 6 days of magnetic field exposure. There is no significant difference in chick weight among the four groups ($p>0.05$).

There was no significant effect of ELFMF on body weight on any group in domestic chick (Table 1).

4. Discussion

Mostafa et al. reported that exposure to ELFMF is associated with impairment in discrimination between familiar and novel objects [23]. In contrast, Sienkiewicz et al. found no effect of ELFMF exposure on object discrimination [30]. However, the difference between these two studies is that the exposure time to ELFMF was acute and of short duration (45 min, once only) in Sienkiewicz et al.'s study, whereas in Mostafa et al.'s study rats were exposed chronically for 24 h/day for 1, 2 or 4 weeks. Our results show that exposure to ELFMF 50min/day has no effect on response latency of detour learning but exposure to ELFMF 20 h/day significantly delays the detour learning. The present results, along with the previous studies, suggest that exposure to ELFMF for a short time seems to have no significant effect on detour learning but long time exposure seems to produce an impairment on learning.

Additionally, it is well known that the mechanisms involved in consolidation are largely distinct from processes used to recall memories. The present results show that the domestic chicks which were exposed to ELFMF 20 h/day need two more trails to learn the detour learning task than that exposed to sham 20 h/day. But once the domestic chicks learn the detour learning task they can remember the task very well (there is no significant difference in response latency among trials 3, 4, 5, 6 and 7 in 50 min/day exposure to sham or ELFMF and 20 h/day exposure to sham; there is no significant difference in response latency between trials 6 and 7 in 20 h/day exposure to ELFMF). These data suggest that exposure to ELFMF 20 h/day may not disrupt the processes of recall memories but influence the processes of consolidation.

Results from passive avoidance learning in chicks show that a critical feature of memory consolidation is a requirement for new mRNA and protein synthesis [28]. Magnetic field exposure could affect the CREB DNA binding and the Ca^{2+} signaling system which could alter transcription, translation, gene expression and protein synthesis [20,32]. The finding of the present study is limited to the effect of exposure to ELFMF on detour learning behavior. Further investigations on the cellular and molecular mechanisms which mediate the effect of ELFMF on learning and memory are needed.

Another interesting question that the present study arises is the effect of ELFMF on the early development of embryos

[9,11,14,26]. ELFMF can influence cells proliferation, pregnancy and embryo development in certain conditions [2,3]. 50 Hz electromagnetic fields can induce developmental alterations in preincubated chick embryos [14]. The results from normal and transformed cell lines show that ELFMF influences the control of proliferation [13,19]. The role of ELFMF in the onset of pathologies such as cancer has also been extensively addressed [18,21]. Another reproducible effect of ELFMF is their influence on cells in transitional state [22]. The development of early postnatal growth period is a good model to study the effect of ELFMF on cells under intense transformation [10,31]. In the domestic chick brain, spatial learning and memory is under intense growth at around 11 days of age. So this period is a sensitive phase for the development of spatial learning and memory. The cells in the hippocampus are in transitional state during this sensitive phase [6]. Our results show that exposure to ELFMF 20 h/day from 9 days of age to 14 days of age has significant effect on detour learning. The domestic chicks need more time to learn the detour task than the controls. It may be because that the development of spatial learning and memory was delayed by the ELFMF exposure 20 h/day. The present results provide the evidences of the possible cognitive and biological effects of exposure to ELFMF and raise attention to the possible health hazard from constant use of some electric and electronic appliances in everyday life.

Conflict of interest

Our experiments were carried out under careful care of animals and the procedures were approved by the Chinese Academy of Sciences. It is my obligation to meet the requirements about the use of animal on research and all the data presented here were or will not be published elsewhere.

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