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Short communication

# Gender- and age-specific impairment of rat performance in the Morris water maze following prenatal exposure to an MRI magnetic field

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**Abstract**

We examined the effects of prenatal exposure (40 min/day, gestation days 12–18) of rats to a magnetic resonance imaging (MRI) magnetic field (MF) on their performance in the Morris water maze. At 2 months of age, female rats showed impaired performance. The animals spent longer time swimming and used inefficient strategies. However, no significant effects on maze performance were observed at 1 and 5 months of age. No evident maze performance deficit was detected in male rats prenatally exposed to the magnetic field. Thus, we conclude that prenatal exposure to MRI magnetic field induces cognitive/behavioral deficits in female rats at a specific age.


*Theme:* Learning and memory
*Topic:* Behavior

*Keywords:* Prenatal exposure; Magnetic field; Magnetic resonance imaging; Morris water maze; Spatial learning; Sexual difference

There is increasing public concern regarding the safety of exposure to magnetic fields (MFs), including those used for magnetic resonance imaging (MRI) applications. Several lines of scientific research have clearly disclosed a number of potential harmful effects of MFs. For example, exposure to MFs causes deleterious effects on spermatogenesis and embryogenesis in animals, and abortion in both humans and animals [18].

The central nervous system (CNS) is particularly susceptible to MFs, which may markedly affect neural electrophysiological and biochemical activities [16,23]. Exposure to MFs can increase the risk of neurodegenerative diseases in humans [19] and produce deficits in attention, perception and spatial learning in rats [12]. Furthermore, the developing CNS exhibits even higher sensitivity to MFs [6]. Prenatal or perinatal exposure to MFs 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 [17,20]. Therefore, elucidation of the physiological and behavioral consequences of exposure of the developing CNS to MFs requires considerable attention.

MRI is widely used to study fetal development [7,22] and assess brain injuries [1], due to its high capacity of visualizing the brain. The effects of prenatal exposure to MRI on cognitive performance at different stages of postnatal development have not been described in detail. In the present study, we investigate the effects of periodic exposure of pregnant rats to MRI fields on the offspring's performance in the Morris water maze at different postnatal ages.

Fifteen healthy pregnant female Sprague–Dawley rats were used in the present study. Animals were exposed either to MFs or sham conditions between days 12 and 18 of gestation. Eight of the pregnant rats were randomly selected and exposed to the MRI field at 7:00 p.m. for 7 days for 40 min a day. The other seven adult rats were exposed to a sham control field under similar conditions. The MRI field was a 0.35 T 1 kW power impulse magnetic field produced by a

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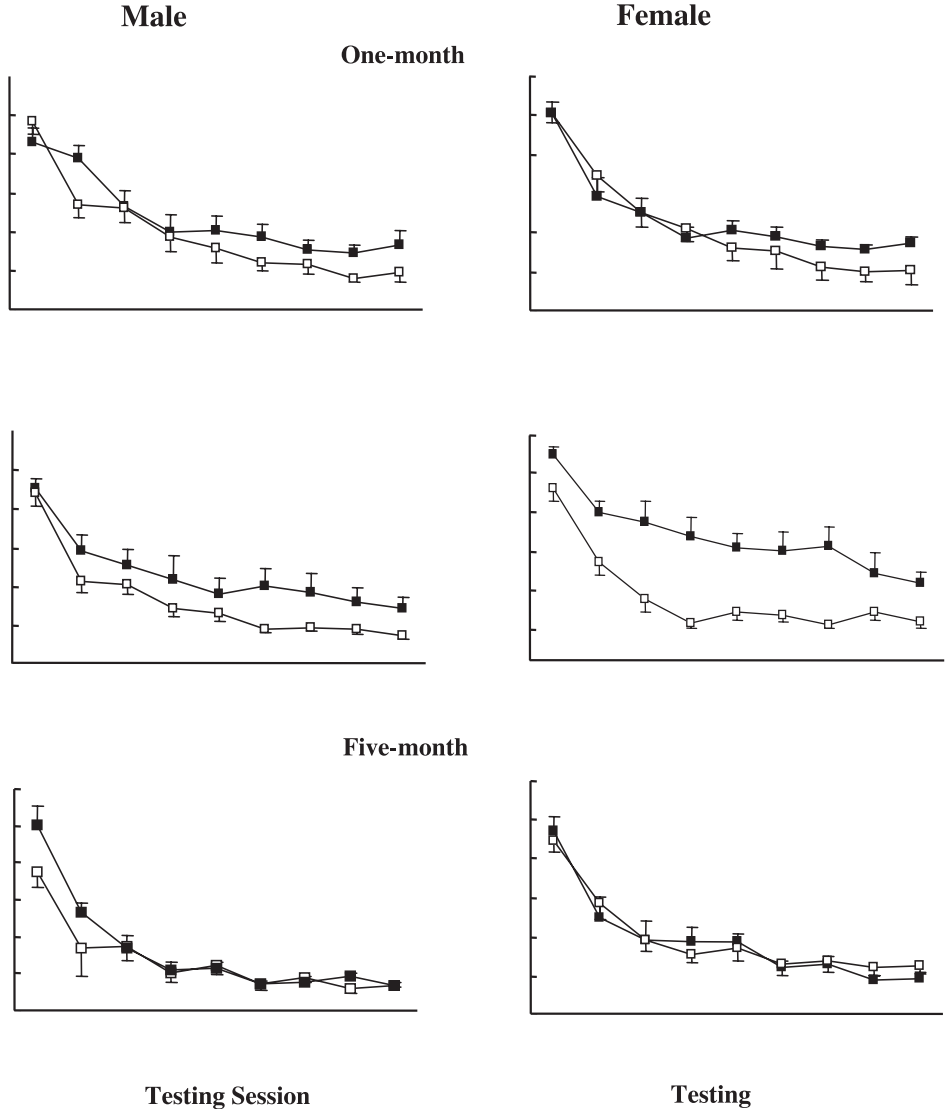
Diasonics MRT-35A system (Comp, USA; spin echo $T_1$: TR 500 ms/TE 15 ms, slice thinness 10 mm, Nex 4; $T_2$: TR 2000 ms/TE 120 ms, Slice thinness 10 mm, Nex 2).

After parturition, each of the 15 litters was housed in a standard plastic cage ($46 \times 31.5 \times 20$ cm) with food and water available ad libitum for the maternal rats. The room temperature was maintained at $21 \pm 1$ °C. The light–dark cycle was 12:12 h (light on at 7:00 a.m.). On the second postnatal day, the number of pups in each litter was randomly reduced to 8. At 21–days of age, offspring were separated from their respective maternal rats and assigned into four general groups: (1) MRI male, (2) MRI female, (3) control male, and (4) control female. Within each group, rats were further randomly divided into three testing-age sets (designated according to their ages when tests were conducted) of 1-, 2-, and 5-month groups. In total, 114 offspring rats were assigned into 12 ($4 \times 3$) groups, and a maximum of two males/females per litter were distributed to each group. The MRI 5-month male group included 8 rats and the control 5-month male group comprised of 6 rats. All other groups contained 10 rats each.

Behavioral tests were conducted using a Morris water maze (120 cm in diameter and 55 cm in height) in a dim room illuminated by a carefully positioned lamp whose reflections from pool water were not perceived by the video camera. The pool was filled with water to a depth of 41 cm, with a temperature of $23 \pm 1$ °C. The position and orientation of the pool in the testing room remained unchanged throughout the study, and thus, both geometric and landmark cues were maintained constant [21]. The pool was divided into four quadrants. A round transparent platform (8 cm in diameter) was placed at the center of a designated quadrant and submerged 1 cm beneath the water surface. The distance from the platform center to the pool edge was 35 cm. The escape latency during



ly towards the platform. In the last test, the time that the rat spent within the ''platform'' quadrant was additionally recorded.

Repeated-measure analysis of variance was applied across sessions. Treatment, age, and sex were the three between-subject factors. Swimming strategies were analyzed with the Mann–Whitney non-parametric test. The time spent on the ''platform'' quadrant in the last test for each rat was analyzed with one-way analysis of variance.

Latencies across sessions for all the animal groups are shown in Fig. 1. All rats improved in performance during the tests, as indicated by the progressive reduction in escaping latencies over the sessions ($F_{8,\ 816}=244.84$, $p<0.05$). For the three between-subject factors (treatment, age, and sex), the interaction of treatment × age was significant ($F_{2,102}=7.501$; $p<0.05$). However, interactions of treatment × sex, sex × age, and treatment × age × sex were not significant.

To analyze the treatment main effects, we examined the differences between control and treatment groups for each age and sex combination. Interactions between testing sessions and treatment were not significant. The treatment main effect was significant for the 2-month female group ($F_{1,18}=17.35$, $p<0.05$), but not for the other groups.

Examination of the strategy profiles used by each animal group revealed that the 2-month-old MRI female rats used marginal strategy more frequently but less tendency and straight strategies than the 2-month-old control female rats. No significant treatment effects were observed in the other groups (Fig. 2).

In the last test, no differences in time spent in the ''platform'' quadrant were observed between groups (Table 1).

The present data show that prenatal exposure to the MRI field resulted in impairment of the Morris water maze performance in 2-month-old female rats only. These females used inefficient strategies and spent longer times to find the platform, compared to their control counterparts. No significant effects were observed in the male groups. These results are similar to previous reports that prenatal exposure to alcohol, toluene, or lead caused poor performance in the water maze only in female offspring of rats [4,10,11], indicating an important sexual difference in response to harmful prenatal environments. It is proposed that males generally use a single type of cues (geometric) in spatial learning, while females depend on multiple cue types [21]. Thus, strategies used by females are considered more complex than the one used by males and therefore may demand more on various signaling processes, causing the females' water maze performance more susceptible to prenatal harmful influences. The present finding, that prenatal MRI exposure has a particularly marked effect on the cognitive performance of 2-month-old female offspring, is in agreement with this theory. This disruptive effect is not apparent in younger or adult females. In view of the previous findings that perinatal exposure to MFs affects sexually dimorphic structures [17], it is of interest to determine whether this sex/age-specific effect is related to the brain sexual differentiation. A number of studies have showed that the sex difference in the midsagittal area of the adult rat *corpus callosum* is mediated, in part, by gonadal steroids in early development, with the sensitive period of hormone action in the female between 25 and 78 postnatal days [3,14]. Additionally, sex differences are evident in spatial learning during early development [5]. The prolonged sexual maturation in females may lead to higher sensitivity to prenatal MFs exposure, and the consequences are more obvious when females have just become sexually mature. Accordingly, the influence of prenatal exposure to MFs on sexual hormonal systems, such as those related to estrogen, should be thoroughly investigated. It is established that estrogen is closely associated with the neural plasticity that occurs during learning [15].

Interestingly, prenatal exposure to MFs is accompanied by an increase in dopamine levels in the offspring's striatum between postnatal weeks 4 and 12 [13]. Estrogen modulates dopamine activity differentially in the striatum of females and males [2], and dopamine activity in the striatum is closely related to the water maze performance [8,9]. Thus, impaired water maze performance in the 2-month-old female offspring suggests an altered estrogen-dopamine interaction pattern in the striatum, following prenatal exposure to the MRI field.

On the other hand, prenatal exposure to the MRI field does not affect the birth weight and litter size of the offspring (data not shown). In our experiments, prenatal exposure was not too severe, and the observed effects in the 2-month female group were not due to a change in birth weight.

The results of the present study strongly call for serious caution in regard to prolonged exposure of pregnant humans to medical MFs. The effects of prenatal exposure to MFs on the interplay between brain sexual differentiation, sexual hormonal modulation, and dopamine activity require further investigation.

Table 1
Average time spent in the platform quadrant in the Morris maze probe tests

| Age | Sex | Group | *N* | Time (s) |
|---|---|---|---|---|
| 1 month | M | CON | 10 | 38.51 ± 2.75 |
| | | MRI | 10 | 37.10 ± 1.94 |
| | F | CON | 10 | 39.36 ± 3.52 |
| | | MRI | 10 | 38.57 ± 2.73 |
| 2 month | M | CON | 10 | 35.24 ± 3.55 |
| | | MRI | 10 | 36.96 ± 3.34 |
| | F | CON | 10 | 34.39 ± 5.19 |
| | | MRI | 10 | 40.13 ± 3.96 |
| 5 month | M | CON | 6 | 37.65 ± 3.78 |
| | | MRI | 8 | 43.93 ± 3.06 |
| | F | CON | 10 | 39.88 ± 2.02 |
| | | MRI | 10 | 40.88 ± 3.09 |

Means ± S.E.M. are presented for each group.

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