ORIGINAL ARTICLE

Early exposure to noise followed by predator stress in adulthood impairs the rat's re-learning flexibility in Radial Arm Water Maze

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Abstract

OBJECTIVE: This study investigated the cognitive effect of chronic exposure to environmental noise on RAWM performance of juvenile rats, and the ability of adult rats exposed to a novel acute stress to perform in the RAWM as a function of whether or not they were exposed to environmental noise as juveniles. **METHODS:** We examined the consequences of exposure to noise during the juvenile-early periadolescent period on adulthood stress response by assessing cognitive performance in the RAWM. Male rats were exposed to environmental noise during the childhood-prepubescent period (21–35 PND), and their RAWM performance was tested at the end of the exposure to noise, and then again two months later when they had to cope with a new stressful event. RAWM execution included a 3-day training phase and a reversal learning phase on day 4. Escape latency, reference memory errors and working memory errors were compared between experimental and control groups. In addition, body weight gain and serum corticosterone levels were evaluated. **RESULTS:** Stressed rats demonstrated spatial impairment, as evidenced by poor execution on day 4. This effect was significantly noticeable in the doubly stressed group. Noise annoyance was evidenced by reduced body weight gain and increased serum corticosterone levels. **CONCLUSIONS:** Our results suggest that environmental noise may produce potent stress-like effects in developing subjects that can persist into adulthood, affecting spatial learning abilities. This cognitive impairment may restrict the subject's ability to learn under a new spatial configuration.

Abbreviations:
RAWM - Radial Arm Water Maze
PND - postnatal day
CORT - corticosterone
HPA - Hypothalamic-Pituitary-Adrenal system

INTRODUCTION

Early-life experience plays a critical role in subsequent neural, endocrine and cognitive development (Hofer 1994; Lewis 2004; Shors 2006). Development therefore represents a critical period for shaping adult behavior. A large body of evidence links early-life stress and a wide variety of alterations in specific brain systems implicated in mature cognitive functions (Weller *et al.* 1988; Lemaire *et al.* 2000; Weinstock 2001; Welberg & Seckl 2001; Maccari *et al.* 2003; Shors 2006; Rice *et al.* 2008; Weinstock 2008). Experimental studies examining the long-term impact of early life experience usually focus on two main stages: gestational stress (Owen *et al.* 2005; Weinstock 2008) and early postnatal adversities, including mother-offspring manipulation during the pre-weaning period (Heim and Nemeroff 1999; Brunson *et al.* 2005; Tang *et al.* 2006). Only recently has a later developmental period, the juvenile or prepubescent stage, been focused upon. Exposure to stress during this period may account for delayed effects on cognitive skills (Maslova *et al.* 2002; Isgor *et al.* 2004; Avital & Richter-Levin 2005; Pohl *et al.* 2007; Toledo-Rodriguez & Sandi 2007). It has been suggested that the time window between weaning and rat adolescence represents an informative model for the study of human childhood because of the increased independence, social behavior, and immature sexuality described in prepubescent rats (Spear 2000, Avital & Richter-Levin 2005; McCormick & Mathews 2007). Moreover, it is known that the hippocampus, a brain structure particularly relevant for learning and memory, continues to grow in the rat long after the first few weeks of life into young adulthood, and that this growth is vulnerable to chronic stress (Isgor *et al.* 2004).

Chronic stress in modern societies is in large part provided by environmental factors. Noise pollution, due to its disturbing nature, has been recognized as a key factor for the development of stress-related diseases (Maschke *et al.* 2000). The progressive growth of noise load in the surroundings of daily life has gained increased scientific attention. Artificial noise models (pure tones or broad-band noise) have been used to investigate the deleterious effects of noise on behavioral and neuroendocrine profiles (Kim *et al.* 2006; Manikandan *et al.* 2006; Samson *et al.* 2007). Rabat and coworkers developed novel adjustments of environmental noise models to fit the rat's audiogram, successfully demonstrating deleterious effects on sleep and memory (Rabat *et al.* 2005; 2006). In this way, environmental noise represents a reliable and relatively unexplored source of stress, the deleterious effects of which can be accentuated by exposure during early development.

Cognitive tasks involving the use of spatial learning are selectively affected by chronic stress (for review see Sandi & Pinelo-Nava 2007). It is believed that the chronic rise in glucocorticoid hormone levels produced by stress mediates this effect (McEwen *et al.* 1986; Oitzl & de Kloet 1992; Oitzl *et al.* 1994; de Kloet *et al.* 1999; Sandi 2003). Moreover, it has been proven that HPA-axis activity can be reprogrammed by chronic exposure to stress during the fetal stage or during the first few weeks of life, permanently changing the endocrine response to stress (Edwards & Burnham 2001, Aisa *et al.* 2007; McPherson *et al.* 2007). Such hormone deregulation induced by adverse conditions during early postnatal life may be highly detrimental for cognitive function and the responsiveness of the HPA axis to subsequent stress (Owen *et al.* 2005; Slotten *et al.* 2006; Meaney *et al.* 2007). Spatial learning and memory are therefore sensible indexes for measuring both short-term and long-term noise stress annoyance. Spatial learning and memory deficits can be studied in spatial maze tasks. The Radial Arm Water Maze (RAWM) task combines the two most widely-used tests assessing spatial learning processes (the radial arm maze and the Morris water maze), and offers a significant advantage because of its ability to simultaneously measuring spatial working memory and reference memory errors (Shukitt-Hale *et al.* 2004).

While very little is known about the cognitive effects of environmental exposure to noise in developing subjects, less is known about the impact of these experiences on the adult response to a novel stressing event. The aims of the present study were thus to determine (i) a possible immediate or delayed effect on RAWM performance of chronic exposure to environmental noise during the postweaning-prepubescent period, and (ii) the ability of adult rats exposed to a novel acute stress to perform in the RAWM as a function of whether or not they were exposed to environmental noise early in their lives.

MATERIALS AND METHODS

Animals

The subjects were 84 Swiss Wistar male rats randomly obtained from an in-house breeding facility at Centro de Investigacion Biomedica de Occidente, Guadalajara, Mexico. The rats were weaned on postnatal day 21, housed in standard polycarbonate cages and maintained on a 12-h light-dark cycle, with lights on at 07:00. Standard Purina rat chow pellets and tap water were provided *ad libitum*. Experimental procedures were approved by the institutional ethics commission and according to the US National Institute of Health Guide for the Care and Use of Laboratory Animals. Body weight was measured routinely on PND 21, 24, 28, 32, 36, 40 and weekly until postnatal day 90.

Study design

To evaluate (i) the immediate effect of chronic exposure to environmental noise during juvenility, and (ii) the long-term effect of this exposure on coping with stressors in adulthood, two experiments were assembled following the juvenile+adulthood stress formula modified from Avital and Richter-Levin (2005) (Figure 1).

Experiment 1: Effects of chronic noise on WRAM performance during juvenility

Here, we tested the immediate effect of exposure to noise stress during juvenility on RAWM performance. From PND 21 to PND 35, rats were randomly exposed to a chronic noise stress protocol (Environmental Noise, n=14) or not exposed (Control n=14). On PND 36, when the juvenile stress protocol was completed, rats from both groups were evaluated in the RAWM and tested for serum corticosterone (CORT) levels.

Experiment 2: Effects of exposure to noise during juvenility, on coping with stressors in adulthood

Here, we tested the ability of adult rats exposed to a novel acute stress to perform in WRAM, as a function of whether or not they were exposed to environmental noise early in their lives. To this end, four groups were created as follows:

1. Environmental Noise + Acute predator stress (EN+APS): 14 rats exposed to noise stress during juvenility (21–35 PND) and then re-exposed to an acute predator stress paradigm at adulthood (90 PND).
2. Acute predator stress (APS): 14 rats only exposed to an acute predator stress paradigm at adulthood (90 PND).
3. Long-term Environmental noise (LTEN): 14 rats exposed to noise stress during juvenility (21–35 PND) that were left undisturbed until adulthood (90 PND).

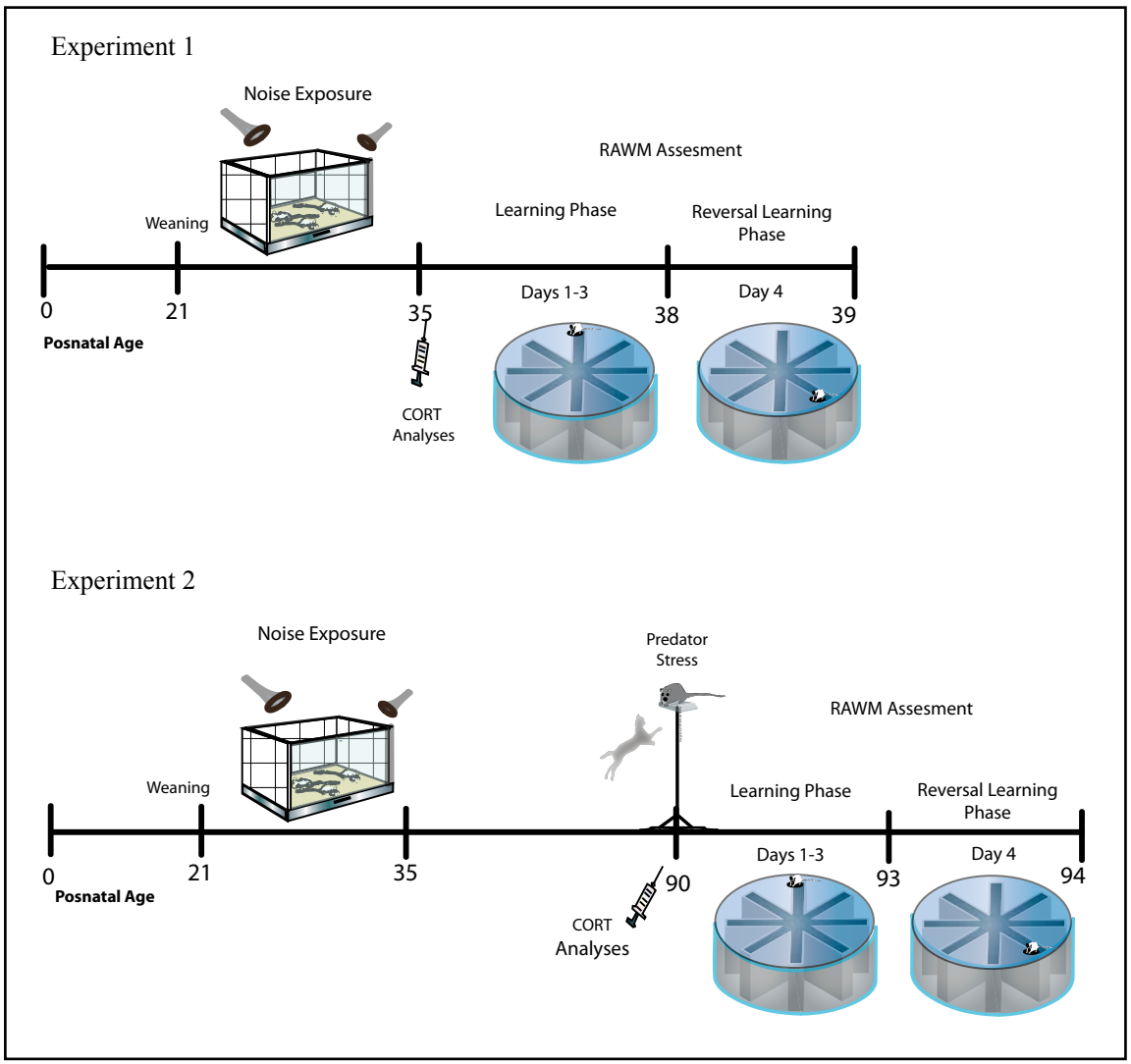

Fig. 1. General procedure. Illustrates the general procedure followed in our experiment. Experimental procedures are chronologically depicted above the line and behavioral assessments (36 and 90 PND) are illustrated below. Environmental noise, acute predator stress and WRAM procedures are schematized.

4. Control: 14 rats that remain undisturbed during all the experiment.

On PND 90, when rats completed the adulthood stress protocol, a new assessment was performed for RAWM and serum CORT levels.

Environmental exposure to noise before the onset of puberty (PND 21–35)

For chronic environmental exposure to noise, the rats' audiogram-fitted adaptation of a noisy environment (Rabat *et al.* 2005) was employed. Briefly, urban audio files containing unpredictable noise events with a duration ranging from 18 to 39 s and spaced by silent intervals ranging from 20 to 165 s were randomly presented to rats during the dark phase (19:00–07:00 h) throughout the 15 days postweaning (21–35 PND) (The audio files were kindly donated by Dr. A. Rabat). Animals were housed in a special sound-isolated acoustic stress chamber provided with professional tweeters (Steren Mexico 80-1088) suspended 60 cm above the solid grid cages and connected to an amplifier (Mackie M1400; freq. 20 Hz–70 kHz; 300 Watts-8 Ω) equipment with mixer software that delivered the acoustic signal at levels ranging from 70 dB for the background noise to 85–103 dB for the noisy events. To make sure that the sound intensity was homogeneous at all places in the cage, noise intensity was measured by placing a Radio Shack Realistic Sound Level Meter (Radio Shack, Mexico) at several locations recording an average of the various readings.

Acute predator stress exposure in adulthood (PND 90)

An inescapable predator stress protocol was carried out by modifying the stress model from Toledo-Rodriguez and Sandi (Toledo-Rodriguez & Sandi 2007). Unavoidable stress was evoked by taking the rat out of the home cage in which it was group-housed, and placing it on an elevated platform (21×20 cm^2, located 90 cm above the ground level) in the middle of a brightly lit room for 30 min. Additionally, an adult cat was transported to the room in a transparent mesh box that allowed the rats access to visual, olfactory, and acoustic stimuli associated with the cat, while preventing physical interaction or attack. The timing of the stress was a single 30 min event between 07:00 and 09:00 on postnatal day 90.

Corticosterone Assays

Immediately after the stress procedure, a subgroup of rats was decapitated and their trunk blood was collected in heparinized tubes. Plasma corticosterone levels were measured using an enzyme immunoassay kit (Correlate-EIA from Assay Designs Inc., USA). Blood samples were obtained immediately after the noise was ended during the juvenile period, or immediately after the rat was removed from the elevated platform during the adulthood experiment (always between 07:00 and 08:00 hours, in order to avoid circadian variation).

Radial Arm Water Maze (RAWM)

Two groups of rats in experiment 1: Control (C) and Environmental Noise (EN), and four groups of rats in experiment 2: Control (C), Environmental Noise + Acute predator stress (EN+APS), Acute predator stress (APS) and Long-term environmental noise (LTEN) were tested for performance in the RAWM to assess spatial learning (Shukitt-Hale *et al.* 2004). In brief, a black round swimming pool (180 cm in diameter, 40 cm height) containing eight radial arms (40 cm high × 43 cm long) was filled with warm water (23±3 °C) to a depth of 32 cm. Fixed to the end of an arm, a submerged platform (10 cm diameter × 30 cm high) was placed, so that it remained hidden from sight. Spatial cues were placed around the pool, which the rat could use to navigate the maze (Figure 1). To begin a trial, a rat was placed in the water and allowed to swim until it found the hidden platform at the end of the goal arm. Each animal did five trials per session (30 min intertrial interval) and the sessions were repeated on four consecutive days. On days 1–3, the hidden platform remained in arm 1 (learning phase), switching to arm 4 on day 4 (reversal learning phase). Each trial started in a different arm and lasted 120 seconds. If a rat could not find the platform was guided to the goal arm and allowed to remain there for 15 s. Then, the animal was removed, dried off, and placed in a holding cage. All these experiments were conducted between 07:00 and 13:00 h on each day. Rats were trained simultaneously on the same day in a closed, sound-proofed room lit with diffuse, attenuated light. All trials were video-recorded to allow manual analysis. We analyzed the following parameters:

Escape latency: It reflects spatial learning and was quantified as the time required to reach the platform.

Reference memory errors: It refers to a memory for information that is held constant over the time and was quantified as the number of entries into arms that never contained the platform.

Working memory errors: It is the ability to remember information that must be distinguished from a similar information previously learned and was quantified as re-entries into an arm (Bizon *et al.* 2009).

Data analysis

The SPSS statistical software was used to perform the statistical analysis. All results are expressed as means ± SEM. A one-way analysis of variance (ANOVA) was run to determine whether noise exposure resulted in differences in body weight gain across the time. Values for CORT and RAWM performance were also analyzed with analyses of variance (ANOVAs) and Tukey´s honestly significant difference (HSD) multiple means comparisons were used to analyze post hoc differences. $p \leq 0.05$ was considered to be statistically significant.

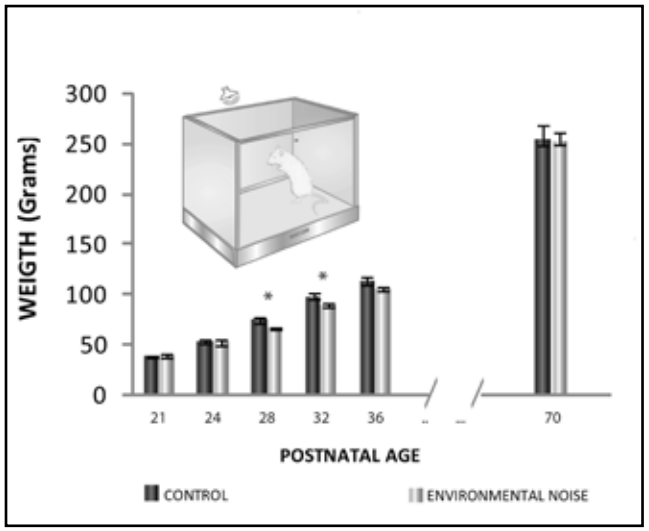

Fig. 2. Body weight gain in environmental noise-exposed rats. The rat's body weight gain expressed in grams from 21 to 70 PND. Bars represent the mean ± S.E.M of the values obtained. Noise-exposed rats gained less weight than control rats (statistically significant only on 28 and 32 PND). *p<0.05.

RESULTS

Effect of environmental noise on body weight gain

Body weight measurements were obtained from rats throughout the experiment (Figure 2). ANOVA revealed that body weight gain was slower in rats exposed to 15 days of environmental noise. Such impairment became statistically significant eight days after the start of the experiment (28 PND) [F(1,17)=6.992; p<0.05] and persisted until the exposure to noise was completed (32 PND) [F(1,17)=5.687; p<0.05]. Once the noise was removed, rats showed a slower body weight gain; however, this trend did not display statistical significance.

Experiment 1: Effects of chronic noise on CORT levels and RAWM performance during juvenility.

CORT response after chronic environmental noise exposure: At 36 PND, CORT serum level was evaluated in rats exposed to 15-day environmental noise and the control group (Figure 3a). We found significant differences for CORT response between these groups [F(1,5)=12.526; p<0.05.

RAWM execution during training period (days 1–3): The control and noise-exposed rats performed a 5-day trial. Results were averaged for simplification and clarity of figures. Statistical analysis revealed no effect on mean escape latency during the days 1 to 3 between the groups. Reference and working memory errors quantification indicated a similar performance for both groups in days 2 and 3. It was only on day 1 that exposed rats showed fewer reference memory errors [F(1,17)=10.118; p<0.01] and working memory errors [F(1,17)=8.123, p<0.05] as compared to controls (left panels of Figure 4b,c).

RAWM execution during the reversal learning (day 4): On reversal learning day, when the hidden platform was changed from arm 1 to arm 4. Animals exposed to 15 days of environmental noise learned the new location significantly slower than rats from the control group, as revealed by the shorter latency prior to finding the hidden platform [F(1,17)=7.079; p<0.05] (Figure 4a, right panel). A similar effect was found on the reversal learning day for the number of reference memory errors (entries into arms that did not have the platform). Environmental noise-exposed rats committed significantly more reference memory errors than the control group [F(1,17)=7.336; p<0.05] (Figure 4b, right panel). Moreover, working memory impairment

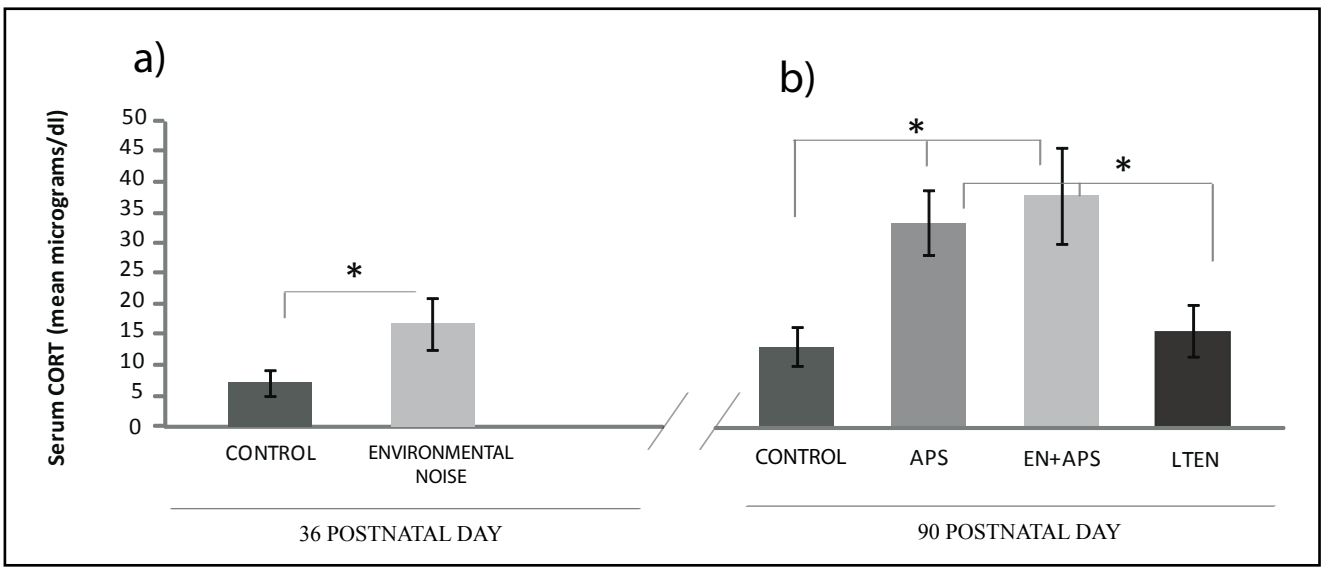

Fig. 3. Serum corticosterone levels in juvenile and adult rats. Mean ± S.E.M plasma levels of corticosterone obtained after exposure to environmental to noise (left panel, 36 PND) and after a new acute stress challenge (right panel, 90 PND). Increased CORT levels were found after exposure to noise (*p<0.05) and immediately after the adult rats were exposed to a novel acute stress, in both the naïve exposed group (*p<0.01) and in the group previously exposed to noise (*p<0.001).

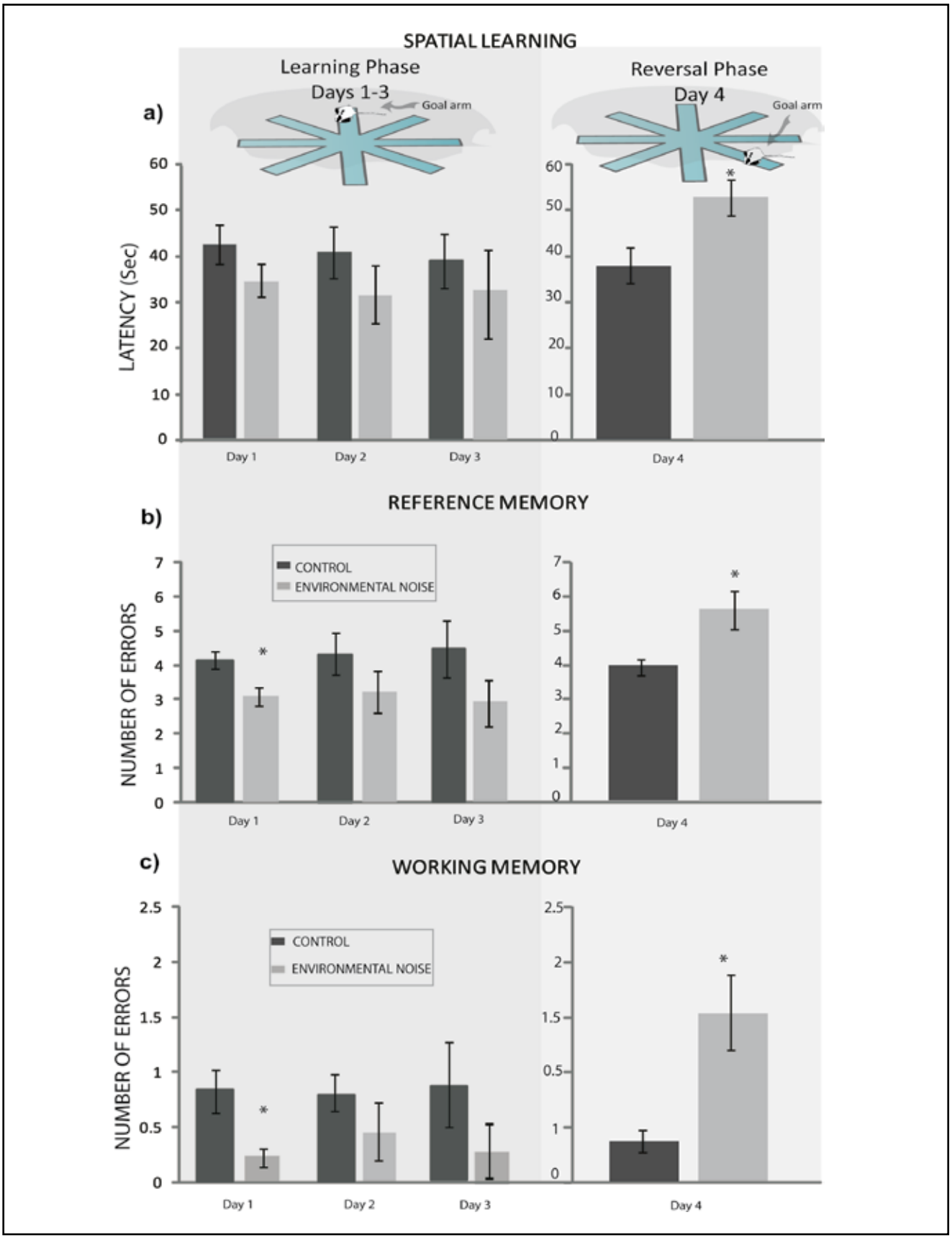

Fig. 4. RAWM performance after juvenile exposure to noise (36 PND): RAMW performance of juvenile rats for the four days following the end of the environmental noise protocol. Left panels show the first 3 days (5 trials per day) of learning assessment when the platform (left top scheme) remained in the same location. Right panels show the fourth training day (5 trials per day) when the platform was removed to another position. Impairment was observed on the reversal day for a) escape latency, b) reference memory errors, and c) working memory errors (*p<0.05).

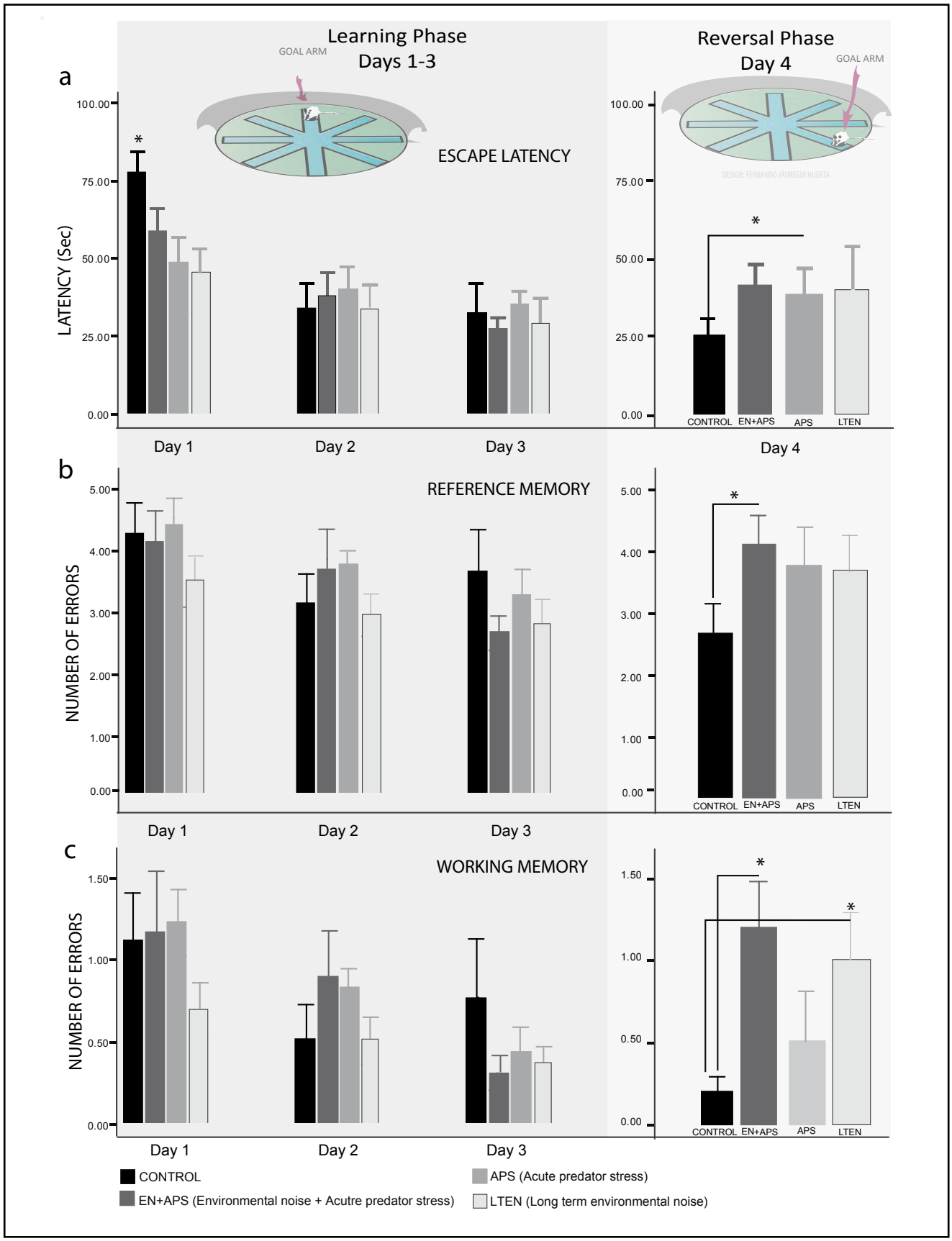

Fig. 5. RAWM performance after a new stress exposure (90 PND). RAWM execution in Control, Acute predator stress (APS), Environmental noise + acute predator stress (EN+APS) and Long-term environmental noise (LTEN) groups evaluated at PND 90. Left panels show the first 3 days (5 trials per day) of learning assessment when the platform (top scheme) remained in the same location. Right panels show the fourth training day (5 trials per day) when the platform was removed to another position. EN+APS group exhibited the most robust impairment on the reversal day in terms of: a) escape latency, b) reference memory errors, and c) working memory errors (*p<0.05).

was a consequence of environmental exposure to noise, as revealed by the increased working memory errors (re-entries into an arm) committed by rats from the exposed group in comparison with values obtained from animal from the control group [F(1,17)=10.580; $p<0.01$] (Figure 4c, right panel).

Experiment 2: Effects of exposure to noise during juvenility, on coping with stressors in adulthood.

CORT response at 90 PND: ANOVA revealed significant differences between the groups for CORT response at 90 PND. Adult rats exposed to acute predator stress (APS) exhibited significantly increased CORT levels as compared to adult controls and the environmental noise (LTEN)-exposed rats (ANOVA-Tukey; $p<0.01$). Accordingly, rats exposed to environmental noise as juveniles and acute predator stress as adults (EN+APS) showed a significant increase in serum CORT levels as compared to the control group and the rats exposed only to environmental noise as juveniles (LTEN) (ANOVA-Tukey; $p<0.01$). No differences were seen between the doubly stressed EN+APS group and the singly stressed APS group, or between the adult rats exposed to environmental noise as juveniles (LTEN) and their corresponding controls (Figure 3b).

RAWM execution during training period (days 1–3): We did not find a main effect of group on escape latency, reference memory errors or working memory errors across the days 1–3. However, in day 1, separate one-way ANOVA revealed reduced latencies from all stressed rats as compared with the control group: EN+APS [F=9.934; p>0.05], APS [F=6.857; $p<0.01$], LTEN [F=8.401; $p<0.01$] (Figure 5a, left panel). At days 2 and 3, no significant differences in escape latency were found among the four groups. No differences were observed on working and reference memory errors assessed at days 1–3 (left panels of Figures 5b,c).

RAWM performance on reversal learning (day 4): At day 4, the escape platform was moved from arm 1 to arm 4, and the animals were given 5 consecutive trials to find the new position of platform (reversal learning). As shown in fig. 5, reversal learning test showed statistically significant differences between controls and juvenile+adulthood stressed rats (EN+APS). Thus, juvenile exposure to environmental noise followed by acute predator stress into adulthood showed a significant impairment in the re-learning ability as indicated by latency measures [F(1,17)=9.934; $p<0.01$]. Moreover, re-stressed rats made considerably more errors during reversal learning, as measured by reference memory errors [F(1,17)=6.932; $p<0.05$] and working memory errors [F(1,17)=14.027; $p<0.01$]. In contrast, rats that were subjected to a single stressful event showed a less severe impairment in their reversal learning ability. Acute predator stress-exposed rats showed impairment in the escape latency [F(1,17)=9.999; $p<0.05$], but not in the reference or working memory errors. The environmental noise-exposed rats that were evaluated at 90 PND (long-term environmental noise group) showed significant impairment only in working memory [F(1,17)=6.345; $p<0.05$]. No statistically significant differences were observed between stressed groups.

DISCUSSION

The results of the present work demonstrate that chronic exposure to environmental noise in the so-called juvenile stage can exert deleterious stress-like effects by reducing the normal body weight gain, increasing the serum corticosterone levels and affecting the rat's ability to perform a spatial learning task in the RAWM. Our results also demonstrate long-term effects of such juvenile exposure that modulate the rat's ability to cope with stress later in life, as was revealed by RAWM performance after exposure to a new acute predator stress. Furthermore, we were able to distinguish a temporal impairment in the reversal learning ability of those rats.

As expected on the basis of results from other authors, we first investigated whether this novel environmental noise protocol induces stress-like effects. We examined whether exposure to stress modulates the rate of body weight gain in the juvenile-exposed rats, and whether this stress increased serum corticosterone levels. Body weight gain reduction was observed on noise exposed rats and CORT levels appeared elevated on PND 36, the day after the last experimental exposure to environmental noise. Thus, classical indicators of stress induced annoyance were found in the noise exposed rats. Although we performed no inter-exposure measurements, we can hypothesize that HPA-axis activity remained elevated during most of the noisy days. This hypothesis may be supported by other reports showing that HPA activity remains altered when an interstimulus interval is introduced to the noise protocol (Atkinson *et al.* 2006; Masini *et al.* 2008). As shown in a previous a study, this stress model did not produce habituation upon a 9-day noise exposure (Rabat *et al.* 2005). Considering that our CORT measurements were recorded after the final exposure to noise, we can assume that a similar or even more pronounced elevation occurred on previous exposure days. Both elevation in glucocorticoid hormones and reduced body weight gain are widely accepted as stress indicators (Herman & Cullinan 1997; Maschke *et al.* 2000; Tsigos & Chrousos 2002).

RAWM performance was affected as a consequence of exposure to environmental noise. When rats had to learn in a new spatial configuration, exposed rats showed cognitive impairment evidenced by a longer time to find the submerged platform on the reversal learning day. In addition, stressed rats committed significantly more errors (both reference and working memory) than rats from the control group. Similar

results were obtained by Shukitt-Hale and coworkers, who assessed the effect of age using the same water radial arm maze protocol. Aged rats showed a similar execution profile with process limited impairment in the reversal learning ability and perseverant behavior (Shukitt-Hale *et al.* 2004). Reversal learning involves a cognitive process that allows the perception and adaptation to new informational cues; such cognitive process could be altered under aging and some pathological circumstances, which cause animals to persistently use behavioral strategies based on old rules that are no longer applicable (Salazar *et al.* 2004). Glucocorticoid receptors could be mainly involved in such impairment because similar effects were observed in dexamethasone treated rats (Cerqueira *et al.* 2005). Moreover, a recent experiment evaluating the effect of chronic-mild-stress on cognition, failed to observe spatial learning impairment in stressed rats, but displayed significant impairment when tested those animals in a reversal learning task (Bessa *et al.* 2009). Thus, reversal learning could be a more sensible process than spatial learning to the aversive effects of mild stressors including environmental noise.

Reversal learning impairment was reanimated by subsequent stress. A primary aim of this study was to assess the ability of adult rats exposed to a novel acute stress to perform in the RAWM, as a function of whether or not they were exposed to environmental noise early in their lives. Again, the most important effect was observed on the fourth day of cognitive assessment when the rats had to learn a new platform location. The juvenile+adulthood stressed group showed the most dramatic and consistent effect. In these rats, detrimental effects on escape latency, reference memory and working memory errors were observed. In contrast, rats that were subjected to a single stressful event showed a less noticeable effect. Acute exposure to inescapable predator stress produced only a significant impairment in escape latency. Long-term effects of juvenile exposure to noise were only evident in the working memory errors. Based upon these results, it appears that both conditions, early exposure to environmental noise and later adulthood acute stress, generated an *additive-like* effect, because neither of these conditions produced an equally detrimental effect by itself. Thus, early exposure affected selectively the late response to acute stress. These results are further supported by previous studies showing that learning and memory processes in adult rats exposed to stress are positively or negatively affected depending on whether they were exposed to a stressor early in life. Acute juvenile stress for example, increased the ability of rats to perform in the water maze after a new stress event, registered 4 weeks later. However, the same task execution was impaired when tested without the novel exposure (Avital *et al.* 2006). Stress differentially affected the acquisition, consolidation and extinction of fear memories as assessed in middle adolescence or adulthood. Males stressed before puberty showed a decreased ability to extinguish auditory fear memories in adulthood (Toledo-Rodriguez & Sandi 2007). Studies that investigate a chronic stress paradigm in the juvenile stage with posterior cognitive assessment, revealed either impairment (Isgor *et al.* 2004) or no effect (Toth *et al.* 2008) on hippocampal-dependent learning. The latter finding (no effect) appears similar to our results if we consider the first three days of training when learning conditions were stable. From this point of view, it is likely that spatial learning is a cognitive process resistant to the long-term effects of the particular stress protocol used in the present work. However, the re-learning capability evaluated on the reversal learning day appears more sensitive to the delayed effects of early exposure to noise. On this line, the reduced flexibility observed again becomes the key finding of this study. This impaired ability to adapt to new informational cues has been described in some stress-related pathologies such as post-traumatic stress disorder and depression (Heim & Nemeroff 1999; Teicher *et al.* 2003). Coincidently, prefrontal cortex is mainly involved in both, behavioral flexibility and stress related disorders (Lapiz-Bluhm *et al.* 2009). Thus, our results support the hypothesis that stress prior to puberty disrupts the normal prefrontal cortex development resulting in abnormal functioning at adulthood (Toledo-Rodriguez & Sandi 2007). In addition, our results supports the hypothesis that juvenile aversive experiences determine future coping ability to stressful conditions; a suggestion that has been recently proposed as the basis of the increased predisposition to anxiety and depression that follows psychology trauma during childhood (Teicher *et al.* 2006; Tsoory *et al.* 2007). Several studies in rats suggest that exposure to stressors during postweaning-periadolescent stages affects limbic system development. Exposure to juvenile stress permanently affects the function of the neural cell adhesion molecule, an essential protein in neural development, regeneration, synaptic plasticity and memory formation processes (Tsoory *et al.* 2008). Furthermore, it has been reported that stress during the peripubertal-juvenile period induces deficits in hippocampal morphology, cognition, HPA-axis function, and brain neurosteroid levels (Isgor *et al.* 2004; Avital *et al.* 2006). Thus, we hypothesized that the long-term effects of early exposure to noise were only evident on the spatial working memory, some endurable changes may occur at behavioral or morphological levels, which allowed the rats to respond with more dramatic impairments when confronted with a new stressing condition. Taken together, noise represents a relevant environmental stressor that contributes to the reduced behavioral flexibility frequently associated with stress-related pathologies.

CONCLUSION

Chronic environmental noise exposure affects the learning capabilities by reducing the juvenile rat's flexibility to learn under a new spatial configuration. Such impairment appears to be permanent enhanced when the rats – as adults – are confronted with a new acute stressing condition. Since studies carried out in the prepubertal period of the rat are considered relevant to conditions in human childhood, attention must be focused on research regarding the particular mechanisms linking these kinds of impairments with some experience-induced predispositions to stress-related psychopathologies.

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