# Impairment in Spatial Memory in adult Rats following developmental Low Lead Exposure

Rajashekar Rao Barkur* and Laxminarayana K. Bairy**
*Department of Biochemistry, Melaka Manipal Medical College, Manipal University, Manipal, India,
**Department of Pharmacology, Kasturba Medical College, Manipal University, Manipal, India
E-mail: rajashekarraob@gmail.com

**ABSTRACT :-**

The present study was aimed to investigate the effect of environmentally relevant levels of lead exposure during gestational and early postnatal period on hippocampal dependent spatial memory in rats during adulthood. The pregnant rats were allowed to drink either normal water (control group) or 0.2% lead acetate solution (Lead treated group) during pregnancy and lactation. Thus rats pups of lead treated group where exposed to lead indirectly through their mothers during this period. At weaning pups of lead treated group were allowed to drink normal water till they attain the adult hood. Blood lead level was estimated on postnatal day 22 and 120. Birth weight and weight gain of the rat pups as they grew were measured at regular intervals. Both the control and lead treated groups of rats were subjected to water maze test on postnatal day 30 and 120. Results showed that lead treatment had no effect on birth weight or weight gain. Blood lead level on postnatal day 22 was significantly high in treated group compared to the control group and it was normalized by end of four months. The rats born to lead treated mothers showed impaired in spatial memory during water maze test both on postnatal day 36 and 126. These data suggests that exposure to environmentally relevant levels of lead during intrauterine and early postnatal period of brain development causes impairment in spatial memory not only during infancy but also lasts till adulthood.

KEY WORDS: Developing brain, hippocampus, lead acetate, Spatial memory, Water maze test

## INTRODUCTION

Lead (Pb) has been long known to be a cumulative neurotoxin (Michael,1998). The lead poisoning in developing countries is due to contamination from industrial wastes, lead battery recycling plants, lead-based paints, silver refining industry, vehicle emission, and folk medicines (Henry,2003).

In humans, first two years of life is considered as the critical period as far as brain development is concerned. In rats, first four weeks of life is considered as crucial period for brain development. During early period brain development, the brain undergoes major developmental and biochemical changes (Rice D,2000). During this period, lead exposure can cause defective brain development and which results in neurological deficit (Rice and Barone.,2000;Evans,2010; Brent *et al.*, 2004). Cookman *et al.*,(1987) have found that lead (Pb) induces precocious differentiation of the glial cells during structuring of the brain during development, further enhancing the chances of alterations in normal brain development.

In humans episodic memory is a type of declarative memory concerned with conscious recall of about facts, events or specific stimuli. It has been proved that the hippocampus contributes to the encoding and storage of episodic memories (Griffiths *et al.*, 1999; Mishkin *et al.*, 1997; Tulving and Markowitsch, 1998). Explicit memory expression is dependent on the hippocampal region in humans (Cohen and Squire, 1980). Hippocampal place cells were suggested as the primary substrate of the spatial memory abilities underlying the spatial navigation processes involved in hidden-platform MWM learning (Poucet, 2000). The hippocampus is critical for spatial cognition in various species of animals, including mice and rats (O'Keefe and Dostrovsky, 1971; Jung and McNaughton, 1993) seems to be the most affected by lead poisoning (Broadbent *et al.*, 2004; Schneider *et al.*, 2001).

The exact mechanism by which lead acts as a neurotoxin remains elusive and lead exposed children show deficits in learning and memory (Chiodo *et al.*, 2007). Experimental data suggests that many neurochemical and neurophysiological processes are affected by lead (Pb) and which result in defective cognitive processes in brain (Finkelstein *et al.*,1998; Johnston and Goldstein *et al.*,1998;). Lanphear *et al.*,(2000) had suggested four affected domains in childhood lead exposure. They are attention, executive function, visual-motor integration and social behavior. However Dietrich *et al.*,(2000) with help of their study suggested that fine-motor coordination and balance are also effected.

Studies conducted in adult rats following developmental lead exposure have shown inconclusive results. Jaako-Movits *et al.*,(2005) found impaired contextual fear conditioning and reduced hippocampal neurogenesis

in adult rat following developmental lead exposure. Murphy and Regan,(1999) in their study conclude that adult rats showed memory deficits following to lead exposure in the early postnatal period. But some researchers did not find any impairment in adult rats' memory retention (Chen *et al.*,1997; Chen *et al.*,2001) and spatial learning (Gilbert *et al.*,2005) following maternal lead exposure.

Even though there are number of studies pointing towards cognitive and behavioral deficits in children who are exposed to lead. The information on long-term consequences in adulthood due to developmental lead exposure is inconsistent. The earlier study conducted in our laboratory found that the passive avoidance learning and memory are affected in adult male rats which were exposed to environmentally relevant lead levels during their embryonic development and early postnatal periods (Barkur *et al.*,2011). So we undertook this study, the goal was to assess the spatial memory during infancy and adult hood (on postnatal day 30 & 120) in rats exposed to low level of lead during their prenatal and early postnatal period of brain development. The spatial memory was assessed by conducting Morris water maze test

## MATERIAL AND METHODS

### Animals

Three months old in-house bred, adult male and female albino Wistar rats were used in this study. Animals were maintained in 12:12 hour light: dark environment, with water and food available ad libitum. Institutional animal ethical committee approval was obtained for the study.

### Experimental design

The lead administration was performed according to the protocol published previously which had produced a blood lead levels between 25 and 35 µg/dl in adult rats maintained on lead (Pb) (Jaako-Movits *et al.*,2005; Chen *et al.*,1997). These blood lead values fall within the environmentally relevant range that found in certain segments of the population throughout the world because of environmental lead (Pb) contamination (Toscano and Guilarte, 2005).The pregnant rats obtained by mating adult female and male rats were divided into two groups. i) Normal control (n=6) and ii) Lead treated (n=6). The rats in the lead treated group were allowed to drink 0.2% (1090 ppm) lead acetate solution through out gestation and for 21 days after delivery (lactation period). Glacial acetic acid (0.5ml/l) was added to the water to prevent any precipitation of lead acetate in the solution. To know the day of conception and gestational age, all female rats were subjected to vaginal smear test. Pups born to lead treated group had access to the same water(water with 0.2% lead acetate) as mother rats until weaning (Postnatal day 21) (Flow chart-1). The rats in the normal control group were allowed to drink normal water through out gestation and lactation period. The male pups born to normal control group of rats and lead treated group of rats were our experimental animals. On 22$^{nd}$ postnatal day, one male rat pups from each of normal control rat (n=6) and one male rat pups from each of lead treated group (n=6) were selected randomly and were housed in separate cages. After wards animals of both groups were allowed to drink normal water till postnatal day 120. The male pups were weighed at birth and at monthly intervals throughout the experimental period. Blood lead level was estimated on postnatal day 22 and 120. The learning and memory retention in male rats of both these groups were assessed by Morris test on postnatal day 30 and 120 (Flow chart-1).

### Blood lead analysis

Blood was collected on postnatal days 22 and 120, from orbital veins in heparinized vacutainers. One hundred microlitres of blood was mixed with 2.9 ml of metexchange reagent using a micro-pipettor and allowed to stand for 24 hours. Blood lead was determined by anodic stripping voltammetry using an ESA-3010B lead analyzer (Kuruvilla *et al.*,2004).

**Morris water maze test** (D'Hooge and De Deyn, 2001)

To test the spatial memory, rats were subjected to Morris water maze test on postnatal day 30 and 120. Data was analyzed using with video camera and Panlab Smart Version 2.5 video tracking software, Barcelona, Spain. The water maze apparatus consists of a water tank of 1.8 meter in diameter, divided into 4 quadrants. There was a 4''x 4'' size escape platform submerged in one of the quadrant, the target quadrant. The pool was filled with water at a temperature of 18-26°C to a depth of about 40cm. Milk was added to the water just before the experiment to make the water opaque. Permanently positioned distinctive objects were placed for facilitating spatial orientation of the animal. Positions of the cues were kept unchanged throughout the period of training. The rats were trained in the water maze in 10 sessions on 5 consecutive days, two sessions on each day. Each session consists of 4 trials. In each trial time taken to reach the hidden platform was recorded. If the rat was unable to find the platform within two minutes, the training session was terminated and a maximum score of two minutes was assigned. Twenty-four hours after the last learning session rats were subjected to memory retention test. This session was of 30 sec. duration. The escape platform was removed from the target quadrant and rat was released in the quadrant opposite to target quadrant. Time taken to reach the target quadrant, time spent in the target quadrant and distance traveled in the target quadrant were measured. Greater latency to reach the target quadrant and less time spent in the target quadrant were suggestive of spatial memory impairment.

**Statistical analysis**

The numerical data are expressed as standard deviation ± SD. Differences between the groups were tested using the unpaired Student's t-test and significance was set at $P<0.05$.

## RESULTS AND DISCUSSION

**Body weight**

The average birth weight and body weights measured at monthly intervals in control and lead-treated pups are shown in Table 1. There was no significance difference between the groups. This indicates that the low level of lead (Pb) dosage (0.2% lead acetate) used in the experiment did not cause any alteration in general health of these animal.

**Blood lead content**

Blood lead content of lead treated group was significantly high compared to normal control when measured on postnatal day 22 (0.23±0.10 µg/dl (n=6) in normal control group vs 31.32±2.76 µg/dl (n=6) in lead treated group)(Figure 1). These blood lead values were consistent with similar data reported by others (12, 14). Blood lead concentration was returned to normal levels in lead treated group by postnatal day 120 as there was no significant difference in blood lead contents between normal control and lead treated group on postnatal day 120 (Figure 1).

**Water maze test on postnatal day 30**

**Latency to escape on to the escape platform during learning sessions.**

In the first session, rats in all groups took more time to reach the escape platform. In the second session rats in all groups were able to reach the escape platform, much faster than 1 $^{st}$ session. In sessions 3, 4, and 5, rats in all groups learnt to reach the escape platform and their escape latency decreased progressively from session to sessions, however lead treated group of rats were taking more time than normal control animals to reach the escape platform. But the data was not significant.

**Water maze test performance in retention test (postnatal day 30)**

**Latency to reach the target quadrant**

The lead treated group of rats took longer time to reach the target quadrant,where as the normal control groups of rats took less time to reach the target quadrant (Figure 2).

**Time spent in the target quadrant (postnatal day 30)**

The lead treated group of rats spent less time in the target quadrant compared to the normal control groups (Figure 3).

**Distance travelled in the target quadrant (postnatal day 30)**

The lead treated group of rats traveled shorter distance in the target quadrant as compared to normal control groups of rats (Figure 4).

**Water maze test on postnatal day 120**

**Latency to escape on to the escape platform during learning sessions.**

The pattern of learning in both rat group was very much similar to the water maze test conducted on postnatal day 30. Lead treated group of rats were taking more time than normal control animals to reach the escape platform during all the 5 learning session, however the data was not significant.

**Water maze test performance in retention test (postnatal day 126)**

**Latency to reach the target quadrant**

The latency to reach the target quadrant was significantly increased in lead treated group of rats compared to the normal control groups of rats (Figure 2).

**Time spent in the target quadrant**

The lead treated group of rats spent less time in the target quadrant as compared to normal control groups of rats (Figure 3).

**Distance travelled in the target quadrant**

The lead treated group of rats traveled less distance in the target quadrant as compared to normal control groups of rats (Figure 4).

Video tracking of water maze test performance of representative rats in each group on postnatal day 36 and 126 is shown in the Figure 5.

## DISCUSSION AND CONCLUSION

The mean blood lead levels in control and lead treated group on postnatal day 22 was 0.23µg/dl and 31.32µg/dl, respectively. On postnatal day 120 blood lead level had come to normal in lead treated group and there was no significant difference in blood lead contents between normal control and lead treated group. So experimental objective of exposing lead (Pb) during crucial period of early brain development in rats was achieved.

The findings from the present study suggest that exposure to lead during crucial period of brain development resulted impairment of spatial memory in this group of rats both at young age and even in adult hood. This can be due to lead-induced impairments of the hippocampus. Hippocampus plays an important role in both spatial and contextual learning. The Morris water maze tests the ability of rodents to learn and memorize the location of a hidden platform in a pool of water by cues keep around water mazes (Morris *et al.*,1982). This task of spatial learning requires involvement of hippocampal NMDA and muscarinic cholinergic receptors (McNamera *et al.*,1993;Ohno *et al.*,1992). It is proved that water maze tasks and passive avoidance tasks in rats require a contribution from hippocampus and other parts of limbic system (Aggleton and Brown,1999;Aggleton *et al.*, 2000).

In this study we observed that lead-exposed rats during learning sessions took little more time to reach the platform on both occasions (Postnatal day 30 and 120) however the data was not significant. However this can be consider as an indication of lead-induced deficit in learning behavior. During the retention test, the lead-exposed rats showed longer latency to reach the target quadrant both during postnatal day 36 and 126. Xu *et al*.,(2009) who exposed rats to similar dosage (0.2% lead acetate) as our study during gestation and lactation found long term memory deficit in Morris water test conducted on postnatal day 30. In our present study we found similar results Morris water maze on postnatal day 36. Xu *et al.*,(2009) also observed damage to mitochondria, microfilaments, and microtubules in hippocampal neurons and myelin sheath degeneration. This might partly explain the reason for the spatial memory deficient in these lead treated rats. There are evidences to suggest that lead exposure causes disturbances in the aminergic system in the cerebral cortex, cerebellum & hippocampus, which contribute to cognitive and behavioral impairments in rats (Devi and Brown, 2005). Jett and Guilarte, (1995) found that developmental lead exposure causes alteration in N-methyl-D-aspartate (NMDA) and muscarinic cholinergic receptors in the hippocampus of rats at 14 days of age. Jaako-Movits *et al.*, (2005) established that exposure to lead during early development inhibits neurogenesis and alters the process of differentiation of new cells in the dentate gyrus of rat hippocampus, which could be responsible for the spatial memory impairments. Studies have also shown that there is decreased adult hippocampal gene expression of nerve growth factor following lead exposure after weaning (Schneider *et al.*,2001) or during lifetime in rats (Cory-Slechta *et al.*,2010) .

Exposure to low levels of inorganic lead during early development has been associated with long-lasting behavioral abnormalities and cognitive deficits in children and young experimental animals (Finkelstein *et al.*,1998; Bellinger *et al.*,1991). In rats exposed to lead from gestation to weaning, Jett *et al.*, (1997) found impairment in performance in the Morris water maze at the age 21days, but not postnatal day 91. In contrast, our study showed spatial learning impairment on both postnatal days 30 and 120. One reason for this discrepancy could be lead doses used. Jett *et al.* (1997) used 250ppm lead acetate in their study which is much less that the dosage used in our study. Neuroanatomical changes caused in brain the by 250ppm of lead acetate might have been reversed by the time animals reached the age of 90 days.

In summary, the results of the present study demonstrate that low-level of environmentally relevant lead exposure during the critical period of early brain development affects spatial memory which lasts a life time.

## REFERENCES

[1] Aggleton, J.P., and Brown, M.W. Episodic memory, amnesia, and the hippocampal-anterior thalamic axis. Behav Brain Sc., 1999,22:425-89

[2] Aggleton, J.P., Vann, S.D., Oswald, C.J., and Good. M. Identifying cortical inputs to the rat hippocampus that subserve allocentric spatial processes: a simple problem with a complex answer. Hippocampus., 2000,10:466-74.

[3] Barkur, R.R., Rao, M.S., and Bairy, L.K. Low Lead Impairs Passive Avoidance Learning in Rats. Archives of Industrial Hygiene and Toxicology., 2011, Jun 1;62(2):147-53.

[4] Bellinger, D., Sloman, J., Leviton, A., Rabinowitz, M., Needleman, H.L., Waternaux, C. Low-level lead exposure and children's cognitive function in the preschool years. Pediatrics., 1991,87:219-27.

[5] Brent, R.L., Tanski, S., Weitzman. M. A Pediatric perspective on the unique vulnerability and resilience of the embryo and the child to environmental toxicants: The importance of rigorous research concerning age and agent. Pediatrics., 2004,113 (Suppl 4):935-44.
Broadbent, N.J., Squire, L.R., Clark, R.E. Spatial memory, recognition memory, and the hippocampus. Proc Natl Acad Sci USA.,2004,101:1451520.

[6] Chen, H.H., Ma, T., Ho, I.K. Effects of developmental lead exposure on inhibitory avoidance learning and glutamate receptors in rats. Environ Toxicol Pharmacol.,2001,9:185-91.

[7] Chen, H.H., Ma, T., Paul, I.A., Spencer, J.L., Ho, I.K. Developmental lead exposure and two-way active avoidance training alter the distribution of protein kinase C activity in the rat hippocampus. Neurochem Res., 1997,22:1119-25.

[8] Chiodo, L.M., Covington, C., Sokol, R.J., Hannigan, J.H., Jannise, J., Ager, J., Greenwald, M., Delaney-Black, V. Blood lead levels and specific attention effects in young children. Neurotoxicol Teratol.,2007,29:53846.

[9] Cohen, N.J., and Squire, L.R. Preserved learning and retention of pattern analyzing skill in amnesia: Dissociation of knowing how and knowing that. Science., 1980,210:207–209.

[10] Cookman, G.R., King, W., and Regan, C.M. Chronic low-level lead exposure impairs embryonic to adult conversion of the neural adhesion molecule. J Neurochem.,1987, 49:399-403.

[11] Cory-Slechta, D.A., Stern, S., Weston, D., Allen, J.L., Liu, S. Enhanced learning deficits in female rats following life time Pb exposure combined with prenatal stress. Toxicol Sci., 2010,117:427-38.

[12] D'Hooge, R., and De Deyn, P.P. Applications of the Morris water maze in the study of learning and Memory. Brain Research Reviews.,2001,36:60–90.

[13] Devi, C.B., Reddy, G.H., Prasanthi, R.P., Chetty, C.S., and Reddy, G.R. Developmental lead exposure alters mitochondrial monoamine oxidase and synaptosomal catecholamine levels in rat brain. Int J Dev Neurosci., 2005,23:375-81.

[14] Dietrich, K.N., Berger, O.G., and Bhattacharya, A. Symptomatic lead poisoning in infancy: a prospective case analysis.J. Pediatr., 2000,137:568–571.

[15] Evans, H,L,. Cognitive and behavioral manifestations of Lead. In: Nass RD, Frank Y, editors. Cognitive and behavioral abnormalities of pediatric diseases. New York (NY): Oxford University Press., 2010, p. 609-16.

[16] Finkelstein Y, Markowitz ME, Rosen JF. Low-level lead-induced neurotoxicity in children: an update on central nervous system effects. Brain Res Rev.,1998;27:168-76.

[17] Gilbert, M.E., Kelly, M.E., Samsam, T.E., Goodman, J.H. Chronic developmental lead exposure reduces neurogenesis in adult rat hippocampus but does not impair spatial learning. Toxicol Sci., 2005, 86:365-74.

[18] Griffiths, D., Dickinson, A., and Clayton, N. Episodic memory: what can animals remember about their past?. Trends Cogn Sci., 1999, 3:74-80.

[19] Henry, F. International environmental health for the pediatrician: Case study of lead poisoning. Pediatrics., 2003,112:259-64.

[20] Jaako-Movits, K., Zharkovsky, T., Romantchik, O., Jurgenson, M., Merisalu E, Heidmets, L.T., Zharkovsky, A. Developmental lead exposure impairs contextual fear conditioning and reduces adult hippocampal neurogenesis in the rat brain. Int J Dev Neurosci., 2005,23:62735.
Jett, D.A., and Guilarte, T.R. Developmental lead exposure alters N-methyl-D-aspartate and muscarinic cholinergic receptors in the rat hippocampus: an autoradiographic study. Neurotoxicology., 1995,16:7-18.

[21] Jett, D.A., Kuhlmann, A.C., Farmer, S.J., Guilarte, T.R. Age dependent effects of developmental lead exposure on performance in the Morris water maze. Pharmacol Biochem Behav., 1997,57:271-9.

[22] Johnston, M.V., Goldstein, G.W. Selective vulnerability of developing brain to lead. Curr. Opin. Neurol., 1998, 11, 689 – 693.

[23] Jung, M.W., and McNaughton, B.L. Spatial selectivity of unit activity in the hippocampal granular layer. Hippocampus.,1993,3:165-182.

[24] Kuruvilla, A., Pillay, V.V., Venkatesh, T., Adhikari, P., Chakrapani, M., Clark, C.S., D'Souza, H., Menezes, G., Nayak, N., Clark, R., Sinha, S. Portable lead analyzer to locate source of lead. Indian J Pediatr., 200,;71:495-9.

[25] Lanphear. B.P., Dietrich, K., Auinger, P., and Cox, C. Cognitive deficits associated with blood lead concentrations 10 microg/dL in US children and adolescents. Public Health Rep., 2000,115:521–529.

[26] McNamera, R. K.,and Skelton, R. W. The neuropharmacological and neurochemical basis of place learning in the Morris water maze.Brain Res. Rev., 1993,18:33–49.

[27] Michael, R. Historical perspective on lead biokinetic models. Environ Health Perspect.,1998,106 (Suppl 6):1461-465.

[28] Mishkin, M., Suzuki. W.A., Gadian, D.G., and Vargha-Khadem, F. Hierarchical organization of cognitive memory? Phil. Trans. R. Soc. Lond., 1997,356:1461-1467.

[29] Morris, R. G. M., Garrud, P., Rawlins, J. N., O'Keefe. Place naviga-tion impaired in rats with hippocampal lesions. Nature., 1982,297:681–683.

[30] Murphy, K.J., and Regan, C.M. Low-level lead exposure in the early postnatal period results in persisting neuroplastic deficits associated with memory consolidation. J Neurochem., 1999,72:2099-104.

[31] O'Keefe, J., and Dostrovsky, J. The hippocampus as a spatial map. Preliminary evidence from unit activity in the freely-moving rat. Brain Res.,1971, 34:171-175.

[32] Ohno, M., Yamamoto, T., Watanabe, S. Effects of intrahippocampal injection of N -Methyl- d-Aspartate receptor antagonists and scopolamine on working and reference memory assessed in rats by a three-panel runway task. J. Pharm. Exp. Ther.,1992,263:943–950.

[33] Poucet,B.E., Save, P., Lenck-Santini, P. Sensory and memory properties of hippocampal place cells, Rev. Neurosci., 2000,11:95–111.

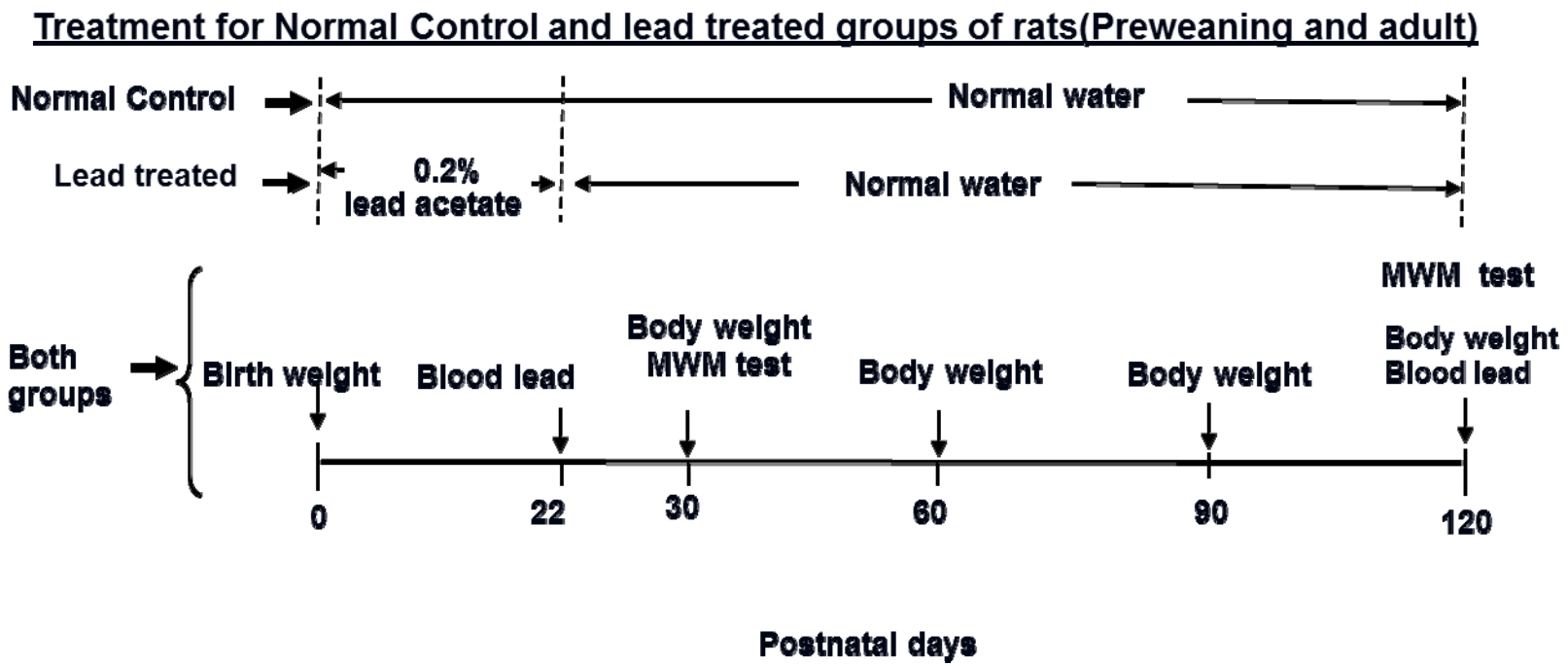
[34] Rice, D., Barone, S, Jr,. Critical periods of vulnerability for the developing nervous system: evidence from humans and animal models. Environ Health Perspect., 2000,108 (Suppl 3):511-33.

[35] Schneider, J.S., Lee, M.H., Anderson, D.W., Zuck, L., Lidsky, T,I. Enriched environment during development is protective against lead-induced neurotoxicity. Brain Res., 2001,896:4855.

[36] Toscano, C.D., and Guilarte, T.R. Lead neurotoxicity: From exposure to molecular effects. Brain Res Rev., 2005, 49:529–554.

[37] Tulving, E., and Markowitsch, H.J. Episodic and declarative memory: a role of the hippocampus. Hippocampus., 1998,8:198-204.

[38] Xu, J., Yan, H.C., Yang, B., Tong, L.S., Zou, Y.X., Tian, Y. Effects of lead exposure on hippocampal metabotropic glutamate receptor subtype 3 and 7 in developmental rats. Journal of Negative Results in BioMedicine., 2009,8:5.




Flow chart-1: Time scale of treatments and measurements for normal control / lead treated groups of experimental rats. MWM- Morris water maze test

| Groups | Body weight (Grams, Mean ± SD) Postnatal day | | | | |
|---|---|---|---|---|---|
| | 0 | 30 | 60 | 90 | 120 |
| Normal control(n=6) | 5.56±0.88 | 51.83±2.71 | 153.17±3.82 | 252.17±2.64 | 289.50±6.47 |
| Lead treated (n=6) | 5.76±0.42 | 53.17±2.12 | 151.50±3.16 | 255.00±3.24 | 287.83±3.56 |

Table 1: Body weight (in grams) of rats in normal control and lead treated groups at different postnatal ages. Each data represents mean ± SD.

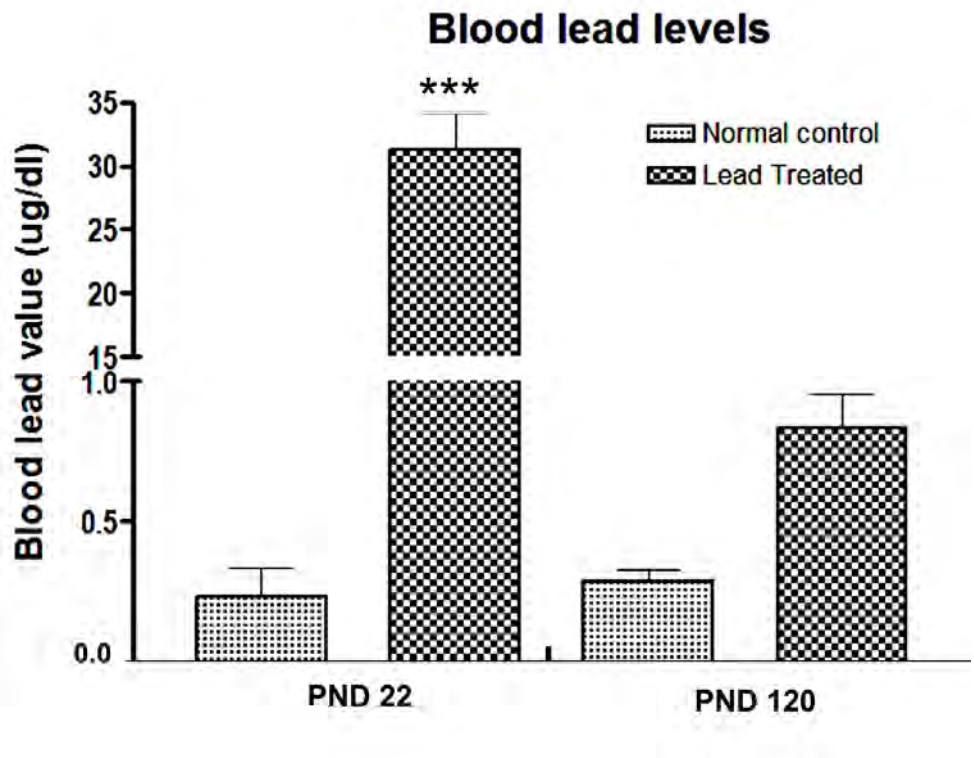



Figure 1

The blood lead content (µg/dl of blood) in normal control and lead treated groups on 22nd and 120th postnatal day (PND). Each bar represents Mean ± SD (n=6 in both the groups). ***P<0.001. (Student's t-test)

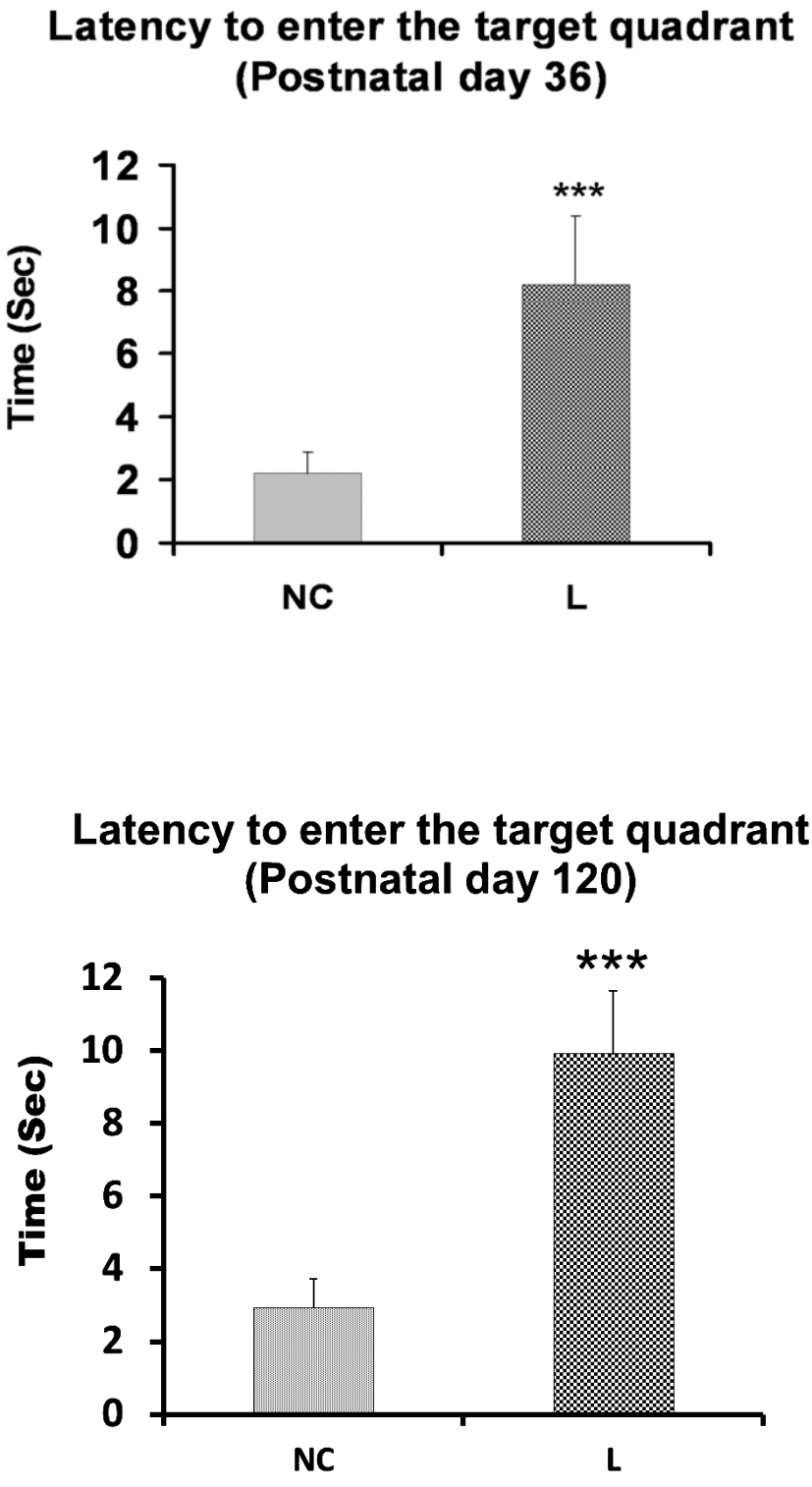



Figure 2

The latency to enter the target quadrant (sec) during the water maze retention test in normal control (NC) and lead treated (L) groups on postnatal day 36 and 126. Each bar represents Mean ± SD (n=6 in both the groups). *** P<0.001. (Student's t-test)

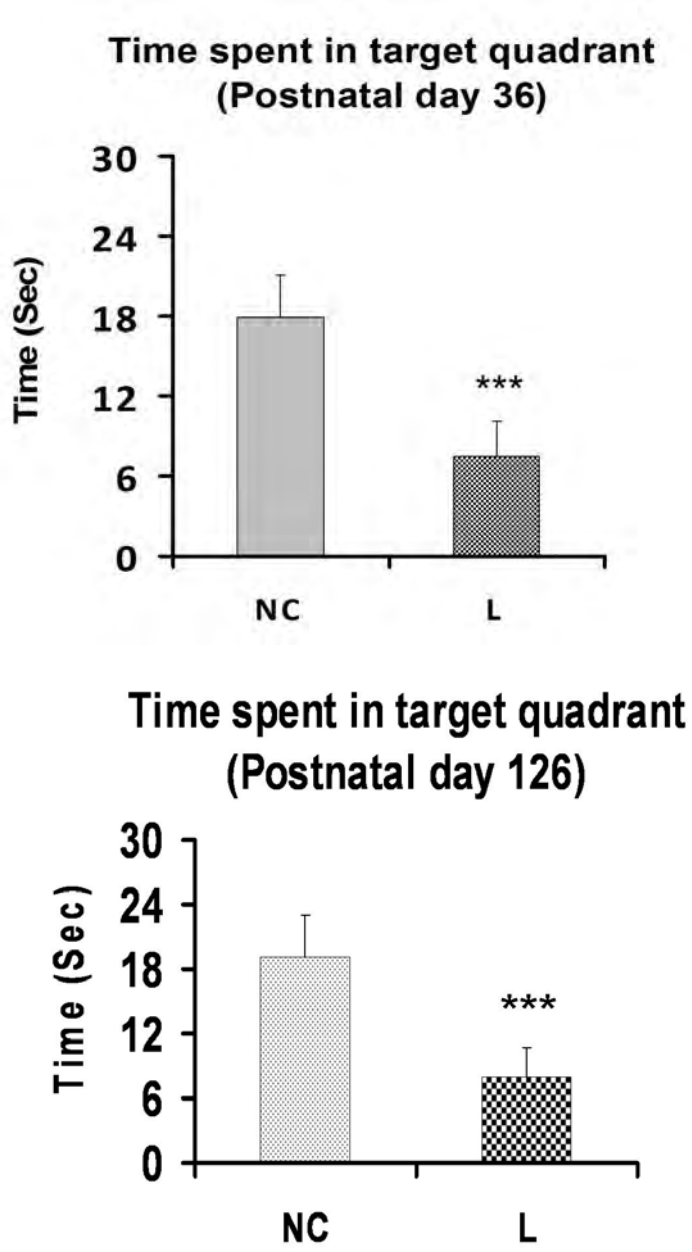
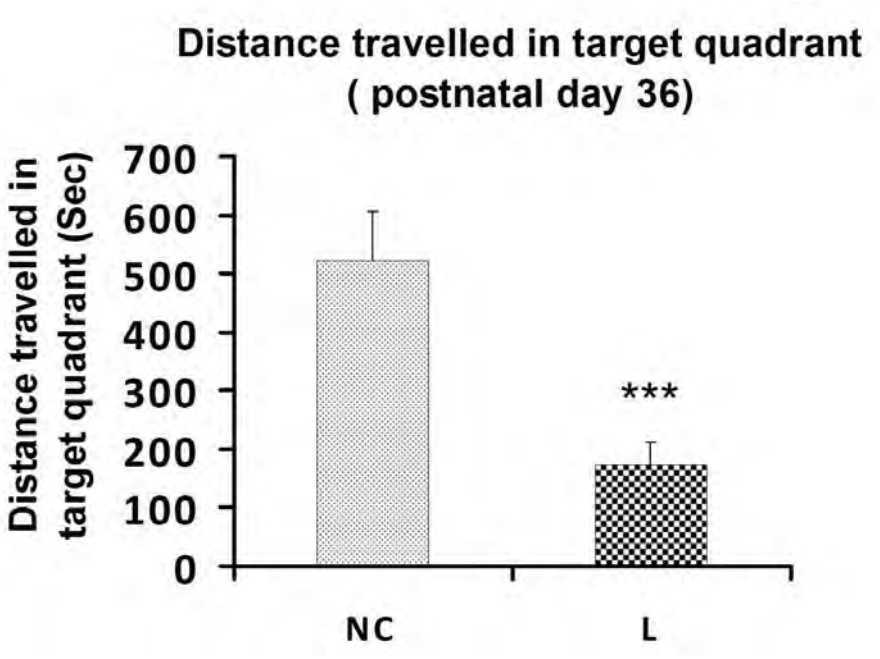



Figure 3

The time spent in the target quadrant (sec) during the water maze retention test in normal control (NC) and lead treated (L) groups on postnatal day 36 and 126. Each bar represents Mean ± SD (n=6 in both the groups). *** P<0.001. (Student's t-test)



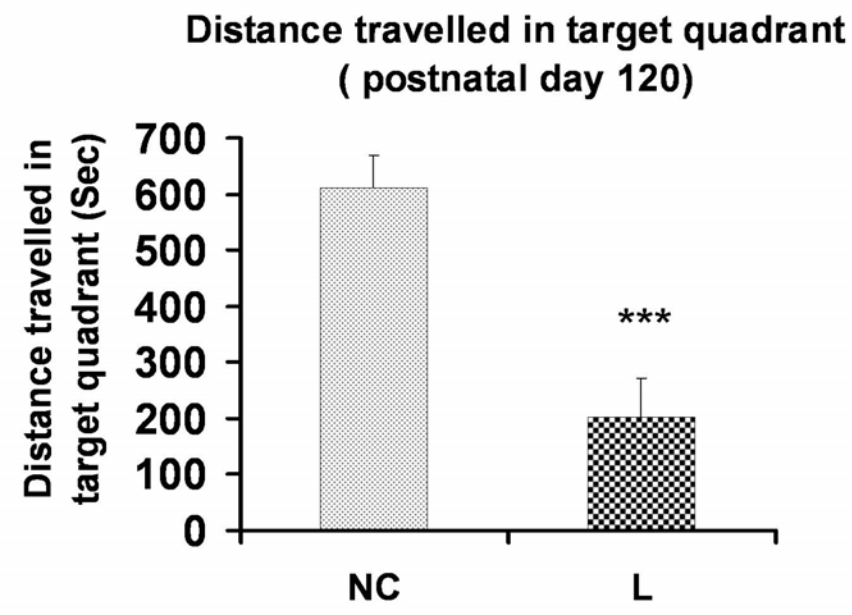




Figure 4

The distance travelled in the target (Sec) during the water maze retention test in normal control (NC) and lead treated (L) groups on postnatal day 36 and 126. Each bar represents Mean ± SD (n=6 in both the groups). *** P<0.001. (Student's t-test)

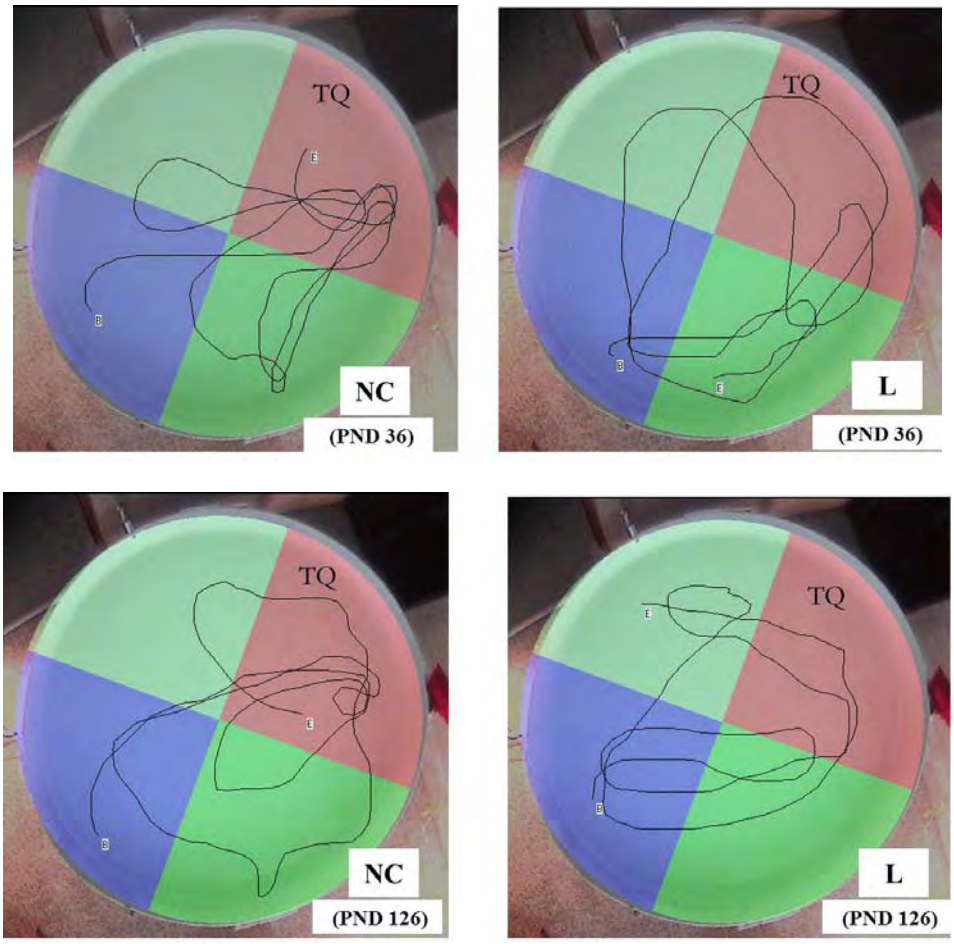



Figure 5

Video tracking of representative rats belonging normal control (NC) and lead treated (L) groups during probe test (spatial memory retention test), 24 hours after last learning session in water maze on postnatal day 36 and 126 respectively. TQ- Target quadrant.