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EFFECTS OF MOBILE RADIATION ON THEMEMORY OF DIFFERENT AGES MALE RATS

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Due to the association of noise pollution and Psychological and physiological diseases, the main aim of this work is to determine the effects of mobile phone radiation on memory of the different ages male rats. In our study male wistar rats were randomly divided into 4 (control and exposed receiving) groups with 10 rats in each. Memory level was measured in animals using eight arm Maze and passive avoidance tests. Data were statistically analyzed where it can be compare between groups using ANOVA and T-Test. The results indicate that memory level was decreased for rats exposed to mobile phone radiation compared to control rats.

1- INTRODUCTION

In physics, electromagnetic radiation (EMR) refers to the waves (or their quanta, photons) propagating (radiating) through space- time frame, carrying electromagnetic radiant energy[1,2].In the range of radiofrequency (RF) electromagnetic waves, it is clear that the mobile phone waves became as an important tool for human activities. An increase use of mobile phones may have a number of physiological and psychological effects on human health. Many studies on animal and human showed various effects on the central nervous system and cognitive performance form of exposure to electromagnetic fields .In this work, we Estimate the effect of Radiation of mobile phones on different age for Male Albino Rats by calculating the mean number of errors in each day, and determining if there is a medical recovery or not from the effect of radiation of mobile phones.There have been a number of scientific reports concerning the possible relationship between exposure to radio frequency fields (RF) during using of mobile phone and reported harmful effects ascribed to regular cell phone usage [3-5]including Headaches, Dizziness, Nausea, Memory loss, Mood rage, Sleep disorders

Fatigue, Loss of concentration, and Lack of coordination,Pain in hands or arms and Skin.The effect was appear even for a 5 minute use of the MP. Working memory deficits are greater not only for the 60 years old and above participants but also for individuals with Mild Cognitive Impairment. These results are in agreement with previous studies on animals as well as humans on the effects of MP use on the brain.

2- MATERIAL AND PROCEDURES

Animals The study includes 80 male Wister albino rats different ages adult and premature.

The rats were maintained under standard laboratory conditions in a room, where the temperature was kept at constant humidity (40-50%), and hold on a 12/12-hour light/dark cycle throughout the experiment. They provided with standard food pellets and water adlibitum. All equalrats were divided into eight groups as follow:

Forty adult group (3 month old) with body weight (220-240 gm) were divided randomly into :

1- Control adult group G1(3 month years old): It included 10 adult male rats (without stress and with the EMR exposure device turned off)

2- Exposed adult group G2: It included30 male rats with the mobile exposure device on it included

* G2 is10 malerats after exposure to 50 missed call for 30 days, symbolically $G2_{50\ A}$.

* G2 is10 male rats after exposure to 5 min call for 30 days, symbolically $G2_{\ 5\ min\ A}$

* G2 is 10 male ratsafter exposure to 10 min call for 30 days, symbolically $G2_{\ 10\ min\ A}$

3- Recovery adult group: It includes the same rats of the exposed adult of the previous group with the mobile exposure device off for the same duration of the exposure of the previous group [7]

* G3 is10 malerats for 30 days without exposureto 50 missed call, symbolically $G3_{50}$

* G3 is 10 male rats for 30 days without exposure to 5 min call, symbolically $G3_{5\ min}$

* G3 is 10 male rats for 30 days without exposure to 10 min call, symbolically $G3_{10\ min}$

Forty premature rats, 45 days old with body weight (140-160 gm) were randomly divided into :

4- Control premature group G4: It includes 10 male rats (without stress and with the mobile exposure device turned off)

5- Exposed premature group (45 day years old): It includes 30 male rats with the mobile exposure device on.

* G5 is10 male rats after exposure to 50 missed call for 30 days, symbolically $G5_{50\ A}$

* G5 is10 male rats after exposure to 5 min call for 30 days, symbolically $G5_{5\ minA}$

*G5 10 is 10 male rats after exposure to 10 min call for 30 days, symbolically $G5_{10\ min\ A}$

6- Recovery premature group : including 30 rats of the Exposed premature group with the mobile exposure device off for the same duration of exposure of the previous group

* G6 is 10 male rats for 30 days without exposure to 50 missed call, symbolically $G6_{50}$

* G6 is10 male rats for 30 days without exposure to 5 min call, symbolically $G6_{5\ min}$

* G6 is10 male rats for 30 days without exposure to 10 min call, symbolically $G6_{10\ min}$

(2-a) Statistical analysis The data were analyzed using unpaired T-test. Two-way ANOVA and student Newman-Keuls Post hoc test were used to examine the effect of electromagnetic waves of mobile radiation on Albion rats in different ways versus control group across the five testing days on the number of working memory errors using Graph Pad Prism data analysis program (GraghPad software, Inc, San Diego, CA, USA). A value of P ≤0.05 was considered statistically significant.[6].This function gives an unpaired two sample Student t test with a confidence interval for the difference between the means.

The unpaired t method tests the null hypothesis that the population means related to two independent, random samples from an approximately normal distribution are equal (Altman, 1991; Armitage and Berry, 1994).

Assuming equal variances, the test statistic is calculated as:[7]

$$T = \frac{\bar{x}_1 - \bar{x}_2}{\sqrt{S^2\left(\frac{1}{n_1}+\frac{1}{n_2}\right)}}$$

Where $s^2 = \frac{\sum_{i=1}^{n_2}(x_i-\bar{x}_1)^2+\sum_{j=1}^{n_2}(x_j-\bar{x}_1)^2}{n_1+n_2-2}$

- where $\bar{x}_1$ and $\bar{x}_2$ are the sample means, s^2 is the pooled sample variance, n_1 and n_2 are the sample sizes and t is a Student quantile with $n_1 + n_{2-2}$ degrees of freedom.. Power is calculated as the power achieved with the given sample sizes and variances for detecting the observed difference between means with a two-sided type I error probability of (100-CI%)% (Dupont, 1990).

The unpaired t test should not be used if there is a significant difference between the variances of the two samples; StatsDirect tests for this and gives appropriate warnings. For the situation of unequal variances, StatsDirect calculates Satterthwaite's approximate t test; a method in the Behrens-Welch family (Armitage and Berry, 1994).

Assuming un-equal variances, the test statistic is calculated as:

$$D = \frac{\bar{x}_1 - \bar{x}_2}{\sqrt{\frac{s_1^2}{n_1} + \frac{s_2^2}{n_2}}} \qquad Df = \frac{\left[\frac{s_1^2}{n_1} + \frac{s_2^2}{n_2}\right]^2}{\frac{(s_1^2/n_1)^2}{n_1 - 1} + \frac{(s_2^2/n_2)^2}{n_2 - 1}}$$

Where $\bar{x}_1 and\ \bar{x}_2$ are the sample means, the value s^2 is the sample variance

$$s_1^2 = \frac{\sum_{i=1}^{n_1}(x_i - \bar{x}_1)^2}{n_1 - 1}, \qquad s_2^2 = \frac{\sum_{j=1}^{n_2}(x_j - \bar{x}_1)^2}{n_2 - 1}$$

n_1 and n_2 are the sample sizes and d is the Behrens-Welch test statistic evaluated as a Student quantile with df freedom using Satterthwaite's approximation.Note that is often more robust to use the nonparametric Mann-Whitney test as an alternative method in the presence of unequal variances.

(2-b)**Newman–Keuls method** : To determine if there is a significant difference between two means with equal sample sizes, the Newman–Keuls method use, a formula that is identical to that one used in Tukey's range test, which calculates the q value by taking the difference between two sample means and dividing it by the standard error:

$$Q = \frac{\bar{x}_A - \bar{x}_B}{\sqrt{\frac{MSE}{n}}} Q = \frac{\bar{x}_A - \bar{x}_B}{\sqrt{\frac{MSE}{2}\left(\frac{1}{n_A} + \frac{1}{n_B}\right)}}$$

where Q represents the student zed range value, where χ_A and χ_B are the largest and smallest sample means within a range, MSE (Mean squared error) represents the error variance taken from the ANOVA table, n_A and $n_B$is the sample size (number of observations within a sample). If comparisons are made with means of unequal sample sizes $n_A \neq n_B$ then the Newman–Keuls formula would be adjusted as follows:

$$MSE = \frac{1}{n}\sum_{i=1}^{n}\left(Y_i - \hat{Y_i}\right)^2$$

where n_A and n_B represent the sample sizes of the two sample means. On both cases, MSE is taken from the ANOVA conducted in the first stage of the analysis.

Once calculated the computed q value can be compared to another q critical value which can be found in a q distribution table based on the significance level ,The error degrees of freedom from the

ANOVA table, and the range of sample means could be tested.. If the computed q value is equal to or greater than the q critical value, then the null hypothesis (H0: $\mu A = \mu B$) for that specific range of means can be rejected[8]As the number of means within a range changes with each successive pair wise comparison, the critical value of the q statistic also changes with each comparison, which makes the Neuman-Keuls method more lenient and hence more powerful than Tukey's range test. Thus, if a pair wise comparison was found to be significantly different using the Newman–Keuls method, it may not necessarily be significantly different when analyzed with Tukey's range test.[9,10]. Conversely, if the pair wise comparison was found not to be significantly different using the Newman–Keuls method, it cannot in any way be significantly different when tested with Tukey's range test.

3- RESULTS

Body weigh; at the end of the experiment, there was no significant difference in final mean body weight among all studied group.

(3-a) Passive avoidance test:

For Adult, The results of rat passive avoidance of adult rats which exposed to 50 missed call for 1 month ,are reported in (Fig.1a). The exposed group shows a non significant difference in the Number of entries to compartment (No.) compared to that control group, rat $G3_{50}$ after one month later without exposure ,The recovery group showed significant shorter latency ($P \leq 0.05$)compared to control group and $G2_{50A}$. The exposed group $G2_{5min}$for5 min. opened call per 1 month, showed non significant difference compared to control, after one month later without

exposure, The recovery group G3 $_{5min}$ shows asignificant shorter latency($P \leq 0.05$) compared to control groupG1, but showed no significant difference compared to G2 $_{5\ min\ A}$ given in(Fig.1b).

The results of **G2$_{10\ min}$** of adult rats which exposed to 10 min. opened call for 1 month showed significant shorter latency to enter to compartment compared to control group ($p \leq 0.05$), after one month later without exposure, The recovery group G3 $_{10\ min}$ showed significant diff. ($P<0.01$) compared to control group, and non significant difference compared to G2 $_{10minA}$as shown in (Fig. 1c)

For Premature:Fig.2a shows the results of rat passive avoidance test of **G5$_{50\ missed}$** of premature rats which exposed to 50 missed call for 1 month. The exposed group shows no significant difference to enter to compartment compared to control group, after one month later without exposureG6 $_{50}$ having a significant shorter latency ($p<0.01$) compared to control groupG4, and significant difference ($P \leq 0.05$) compared to G5$_{50A.}$ The exposed G5 $_{5min}$ to 5 min. opened call for 1 month(Fig.2b)showed significant shorter latency to enter to compartment compared to control($p \leq 0.05$), after one month later without exposure, The recovery group showed significant difference ($P<0.001$) compared to control groupG4 ,and no significant difference compared to G5 $_{10minA}$G5$_{10\ min}$ of premature rats which exposed to 10 min. opened call for 1 month and the recovery group G6 $_{10min}$ showed significant shorter latency to enter to compartment compared to control ($P<0.001$) respectively.

(3-b)Eight-arm radial maze: working memory task :

For Adult,Figure 3ashows the number of working memory errors (numbers of re-entries into baited arms; 4 arms contain food) for the three groups. Over all 5 testing days the mean number of errors was decreases over days in the three groups. The exposed G2 $_{50}$to 50 missed call for one month shows no significant difference appears compared to control group also after one month later without exposure, the recovery group showed significant shorter latency to enter to compartment ($P \leq 0.05$) compared to control group, but showed no significant diff. compared to G2 $_{50A}$.The

exposed group G2 $_{5min}$for 5 min. opened call per one month showed non significant difference compared to control group, G6 $_{5min}$after one month later without exposure, the recovery group showed significant difference ($P \leq 0.05$) compared to control group,(Fig3b).

Fig.3c shows the result of Group G2 $_{10min}$ which exposed to 10 min. opened call for 1 month showed significant shorter latency to enter compartment ($P \leq 0.05$) compared to control group G1, after one month later without exposure, the recovery group G6 $_{10\ min}$ showed higher significant difference ($P<0.01$) compared to control group, G3 showed shorter latency but non significant difference compared to G3 $_{10minA.}$

For Premature: Figure 4a shows the number of working memory errors (numbers of re-entries into baited arms; 4 arms contain food) for the six groups. Over all 5 testing days the mean number of errors was decreased over days in the three groups. The exposed group $G5_{50}$ to 50 missed call for one month showed significant shorter latency ($P \leq 0.05$) compared to control group, after one month later without exposure, the recovery group G6 $_{50}$ showed significant difference ($P \leq 0.05$) compared to control group, and non significant difference compared to$G5_{50A.}$ The exposed group G5 $_{5\ min}$for 5 min. opened call per 1 month showed significant shorter latency to enter compartment ($P \leq 0.05$) compared to control group, after one month later without exposure, the recovery group G6 $_{5min}$ showed significant difference ($P<0.01$) compared to control group, and significant difference ($P \leq 0.05$) compared to $G5_{5min}$.. Group G5 $_{10\ min}$ which exposed to 10 min. opened call for one month showed significant shorter latency to enter compartment ($P \leq 0.05$) compared to control group , after one month later without exposure, the recovery group G6 $_{10min}$ showed significant difference ($P<0.001$) compared to control group, and ($P \leq 0.05$) significant difference compared to G5 $_{10\ min\ A}$.

Passive Test For Adult Figure (1)

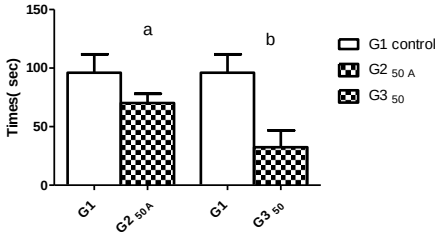

Fig.1 A
Effect of exposure of mobile phone 50 missed call on passive avoidance test in Adult Rats, Data are expressed as mean ± SD
a Non significant as compared with G1 Control
b/ P< 0 .05, G3 $_{50}$ significant as compared with G1 Control
c/ P <0.05 G3 $_{50}$ significant compared with G2 $_{50A}$

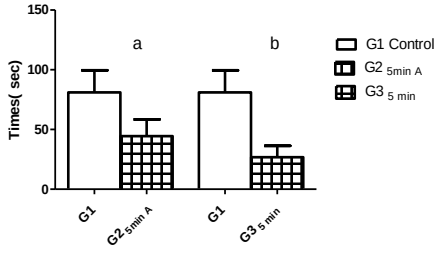

Fig 1B
Effect of exposure of mobile phone 5 min. opened call on passive avoidance test in Adult Rats of G4, Data are expressed as mean ± SD
a : Non significant as compared to control group G1
b /P < 0 .05, G3 $_{5\ min}$ significantly compared to control group G1

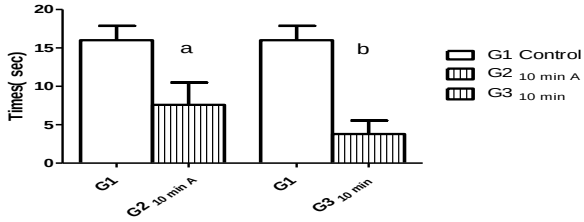

Fig.1C
Effect of exposure of mobile phone 10 min. opened call on passive avoidance test in Adult Rats, Data are expressed as mean ± SD
a / P<0 .05 G2 $_{10\ min\ A}$ is G1 Control
b /P<0 .01,G6 $_{10\ min}$ significantly compared to control group significant compared to control

Passive Test For Premature Figure (2)

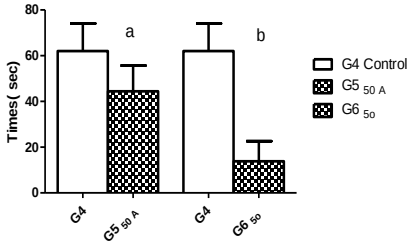

Fig.2A
Effect of exposure of mobile phone 50 missed call on passive avoidance test in Prem. Rats of G6, Data are expressed as mean ± SD
a : non significant compared to Control group G4
b/P<0.01 , G6 $_{50}$ significantly compared to G4
c/ P< 0.05 G6 $_{50}$ significantly compared with G4 $_{50A}$

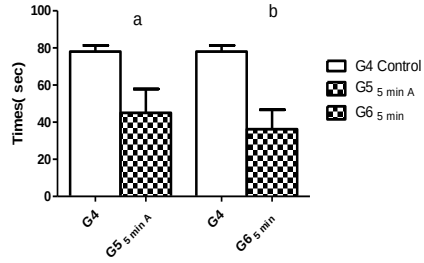

Fig.2B
Effect of exposure of mobile phone 5 min opened call on passive avoidance test in Prem. Rats, Data are expressed as mean ± SD
a/ P<0.05, G5 5 min significantly compared to control group G4
b/ P <0.001, G 6 5 min significantly compared to control group G4

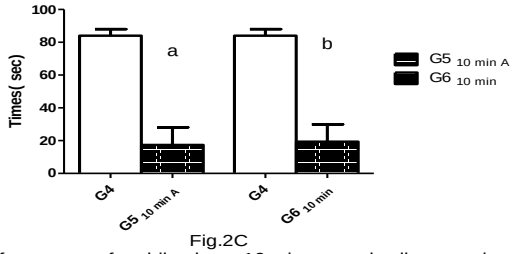

Fig.2C
Effect of exposure of mobile phone 10 min opened call on passive avoidance test in Prem. Rats, Data are expressed as mean ± SD
a/ P<0.001 , G5 $_{10\ min\ A}$ significantly compared to control group G4
b/P< 0.001, G6 $_{10\ min}$ is significantly compared to control group G4

For Adult Maze Test Figure (3)

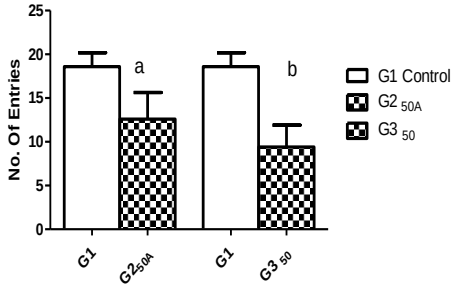

Fig.3A
Radial Maze learning in adult albino Wistar rats exposed to mobile radiation 50 missed call, data represented as mean ±SD
a :G2 50_A is Non significant compared to G1 Control
b / P < 0.05 G3 $_{50}$ significant compared to G1 Control

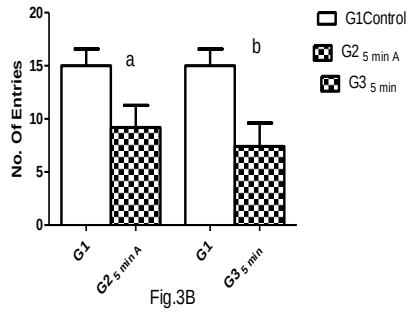

Fig.3B
Radial Maze learning in adult albino Wistar rats exposed to mobile radiation 5 min opened call, data represented as mean ±SD
a :Non significant compared to control group G1
b / p< 0.05 G3 $_5$ min significantly compared to control group

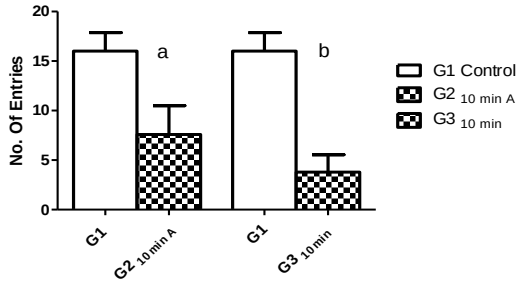

Fig.3C
Radial Maze learning in adult albino Wistar rats exposed to mobile radiation 10 min opened call, data represented as mean ±SD
a/ P<0.05, G2 $_{10}$ min significantly compared to control group G1
b/ P<0.01, G3 $_{10}$ min significantly compared to control group G1

For Premature Maze Test Figure (4)

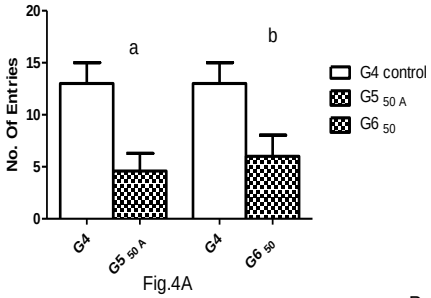

Fig.4A

Radial Maze learning in Premature albino Wistar rats exposed to mobile radiation 50 missed call, data represented as mean ±SD
a/ P< 0.05, G5 $_{50\ A}$ significantly compared to control group G4
b / P< 0.05, G6 $_{50}$ significantly compared to control group G4

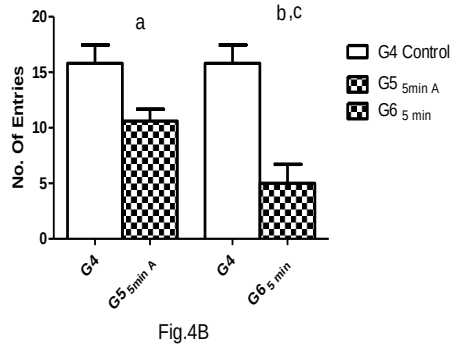

Fig.4B

Radial Maze learning in Premature albino Wistar rats exposed to mobile radiation 5 min opened call, data represented as mean ±SD
a / P<0.05, G5 $_{5\ min}$ A significant compared to control group G4
b/ P<0.001, G6 $_{5\ min}$ significantly compared to control group G4
c/P<0.05, G6 $_{5\ min}$ significant compared to G5 $_{5\ min\ A}$

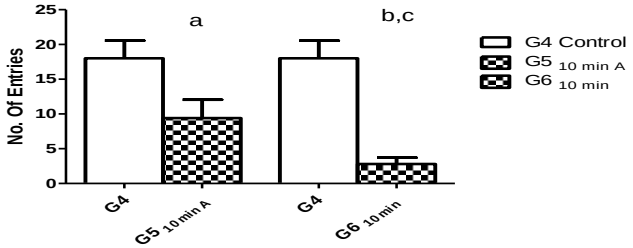

Fig.4C

Radial Maze learning in Premature albino Wistar rats exposed to mobile
radiation 10 min opened call, data represented as mean ±SD
a/ P< 0.05, G5 $_{10\ min}$ A significantly compared to control group
b/ P<0.001, G6 $_{10\ min}$ significantly compared to control group
c/P <0.05 G6 $1_{0\ min}$ significantly compared to G5 $_{10\ min}$

4-DISCUSSION

Our study designed to investigate the short-term effect of EMF (Electromagnetic Frequency)and validated the result with both the behavior test and Passive test.Spatial memory is a type of short-term memory which is responsible for recording surroundings and spatial orientation. Eight-arm radial maze is recognized as an ideal model to study animal spatial memory, which can effectively exclude interference on learning and memory caused by animal's movement dysfunction [11,12].In our study we used adult and premature male rats in different exposure time investigate the effect of mobile phone radiation on memory, each age is divided into 4 groups, control, exposed to 50 missed call, exposed to 10 min opened call\daily and exposed to 5 min opened call \daily for 30 day, and a recovery group, for 1 month after each exposure.

Passive avoidance test rats exposed to 50 missed call for adult group,our experiment showed that there is nosignificant difference between control group and exposed group to 50 missed call and that findings was assured by Dongmei etal. (2013) [11]it was found that there was mild effect but no significant difference between the control and exposure group in the first 5 weeks. However, in this study the effect became more significant in the range from 5^{th} to 8th weeks.There was significant difference between recovery group and control group. In premature group, which exposed to 50 missed call , there was not significant difference between exposed group and control group in our study but Sareesh etal.(2010) [13]reported that there was significant difference in male rats 8-10 weeks old ,exposed to Global System for Mobile (GSM) (0.9 GHz/1.8 GHz) the difference in outcome is due to the fact that it used a lower age.

r**adial maze:** in adult group, it was not found any significant effect in No. of entries after one month, that was assured by Sienkiewicz et al., (2000)[14] who reported that Mice exposed to gigahertz transverse electromagnetic cell (GTEM) GSM 0.05 W/kg 45'/day has no significant effect .However we find a significant effect after 2 month in adult group.In premature group, which exposed to 50 missed call, there was

significant difference between exposed group and control group that what assured by Sareeshet al. (2009) [15] who investigate the cognitive test for rats by (radial maze test) and reported higher significance between recovery group and control group.

Which probably indicated that the exposure to 50 missed call has a long term effect on premature.

In adult group exposed to 5 min opened call for 1 month , we find that there is no significant difference in time taken to enter the dark compartment between exposure group and control , but there was significant difference after 2 moth between recovery group and control group who tested with **Passive avoidance test**

In premature there was significant difference between exposed group and control group and these effect became more significant after two month in premature group.

These findings were assured by Kalafatakis etal.(2017)[16],who reported the effect of mobile phone use for 5 minutes which cause a significant memory impairment in humans.
For adult rats,**Eight arm maze**, there were exposed to 5 min call for one month, showed no significant difference between control group and exposed group, but significant difference after 2 month.

In premature, there was significant effect in No. of entries after one month and higher effect after 2 month between control and exposed group, there was significant difference between exposed group and recovery group.

in passive avoidance test, for adult rats exposed to 10 min opened call, there was shorter time taken to enter the dark compartment after one month between control group and exposed group and least time after 2 month.In premature group, there was significant difference in time taken to enter the dark compartment between control and exposed group, and still short time after 2 month.

Eight arm maze, for adult rats exposed to 10 min. opened call daily for 1 month, we noticed a significant decrease in No. of entries to enter the right arms after 1 month between control and exposed group and after two month there was more significant decrease in number. of entries

These finding was assured by DongmeiHao et al. (2013) [17]who said there was significant difference in week 4 (p = 0.000), week 5 (p = 0.007) and week 8 (p = 0.049).when rats exposed to EMF for 10 weeks.

In premature, , there was significant decrease in No. of entries after one month and more decrease after 2 month between control and exposed group, there was significant decrease between exposed group and recovery group.Yuhong Li et al. [18] said That study demonstrates that the effects of radiofrequency radiation exposure may be time-dependent. Based on these studies, further investigations of EMR effects on cognitive functions may contribute to our understanding of learning and memory impairments in humans and rats. A number of studies reported impairment of cognitive function after exposure to mobile phone electromagnetic field (MP EMF), while others observed no effect or improved performance. The variance in the results may be attributed to methodological issues.

Therefore, it can be stated that exposure to EMF even in a 4-h exposure, can impair memory consolidation in mice. These findings are in agreement with Jadidi et al. (2007), Trimmel and Schweiger (1998), Lai(1996) and Lai and Carino(1999) about the impairing effects of ELF on cognitive functions. Previous studies mainly focused on cognitive functions specially the memory and learning in different tasks and duration of exposure, so inconsistent results are predicted. Jadidi et al. (2007) provide evidence that exposure to a 50 Hz, 8 mT magnetic field for 20 min impaired consolidation spatial memory using water maze but not the retrieval of learned information. The mechanisms underlying the harmful effects of magnetic field on learning and memory are not known.

The brain cholinergic system plays a crucial role in learning and memory (Whishaw, 1989; Whishaw and Tomie, 1987). It has been shown that exposure to ELF decreased the activities of cholinergic system in the frontal cortex and hippocampus; both regions are involved in memory processing (Lai and Carino, 1999). Thus, one possibility is that the impairment of cognition processing can result from decrement in transmission of cholinergic system. Manikonda, etal. (2007) contend that calcium ion could modify once exposing ELF; the modification of

calcium ion and other factors such as GABA has been illustrated in different studies (Blackman, 2009). These changes and other possible impacts on electroencephalography (EEG) may explain the finding thatbrain and its cognitive operations could be affected by exposure to ELF [19].

In summary, findings indicate that 30-min and 90-min exposure to a 50 Hz 8 mT, EMFs can impair memory consolidation in a passive avoidance task. In agreement with others, our data indicates that exposure to ELF has an impairment effect on learning and memory functions. Therefore, further studies are required to find out the underlying mechanisms.

5-CONCLUSION

In summary, our findings indicate that exposure to 50 missed call, 5 min opened call and 10 min opened call /day for one month can impair memory consolidation in adult and premature rats. In agreement with others, our data indicates that exposure to EMF has an impairment and deleterious effects on memory functions. We found that recovery period for all rats for one month did not improve the memory function of the rats, which suggest that the impairment effects of EMF on memory of the rats is permanent not temporary. so, we recommend further studies to find out the effect of longer recovery period and to examine the underlying mechanisms.

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الملخص العربى

يهدف هذا البحث الى تحديد التاثيرات الناجمة عن اشعاعات الهاتف المحمول على ذاكرة الفئران ذوى الاعمار المتباينة وفى دراستنا تم تقسيم مجموعات فئران التجارب الذكور الى اربعة مجموعات احداهما مرجعية و الثلاثة الباقيات تم تعريضها للاشعاع بعدد عشرة فئران لكل مجموعة وتم قياس مستوى الذاكرة للحيوانات باستخدام المتاهه ذات الثمانية اذرع واختبارات التجنب السلبى ثم تم تحليل النتائج والمقارنه بين المجموعات احصائيا و اظهرت النتايج تناقص الذاكرة لدى الفئات الاصغر سنا بمستوي اعلي من الاكبر سنا مقارنةبالفئة المرجعية