AIM OF THE WORK

This study aimed to investigate the potential MRI sound adverse effects on some biophysical (dielectric properties) and biochemical (blood glucose level, malondialdehyde and superoxide dismutase) parameters in the brain of experimental mice. Exploring if there is a correlation between MRI sound power and the severity of these parameters.

MATERIALS AND METHODS

4.1. Materials

1. Sensitive recording tool, [mp3 recording unit].
2. Super power (***Public Address***) PA amplifier ≈ 40 watt audio-output.
3. Laptop fitted with the specified software for sound analysis.
4. Turbo loudspeaker (≈ 20-40 watt r.m.s.).
5. Matt-lab 2010 calculating programs.

-A homemade anechoic chamber 70 × 70 x 70 cm^3, build according to the standard specification.[10] A loud speaker installed in the chamber, driven by the PA amplifier, which can transmit the recorded specified noise with the required decibel value (see Fig.14).

Fig.(14):The anechoic chamber used for mice exposure to the MRI sound.

4.2.Animals:

Mice: 40 mice weighing 25-30 gm were purchased from the animal house, Medical Research Institute, Alexandria University. The animals were housed in an animal facility under 12:12 hr light dark cycle, temperature of 24 ± 1^{o}C, relative humidity of 55 ± 10% and normal atmospheric pressure. They were provided with pelleted food and filtered tap water ad libitum throughout the experiment. All animal experiments were performed according to the ethical guidelines of the Medical Research Institute Alexandria University Ethical Regulation for conducting research on human subjects and animals.[103]

4.2.1Animal groups:

After 1 week of acclimatization, the animals were randomly divided into four maingroups, each groups contains 10 mice.These groups were exposed to sound noise with different power intensties, inside the anechoic chamber (three chambers were used for each the three noise power). Each group was exposed to noise as follow:

- **Control group (GpA):** consists of 10 normal male mice, were remained in the anechoic chamber for two hours daily for a week without exposure to any noise.
- **GpB1** (10 mice): exposed to sound intensity of 121 dBfor 2 hours daily for a week.
- **GpB2**(10 mice): exposed to sound intensity ofits***actual value*** 100 dB for 2hours daily for a week.
- **GpB3** (10 mice): exposed to sound intensity of 90 dBfor 2 hours daily for a week.

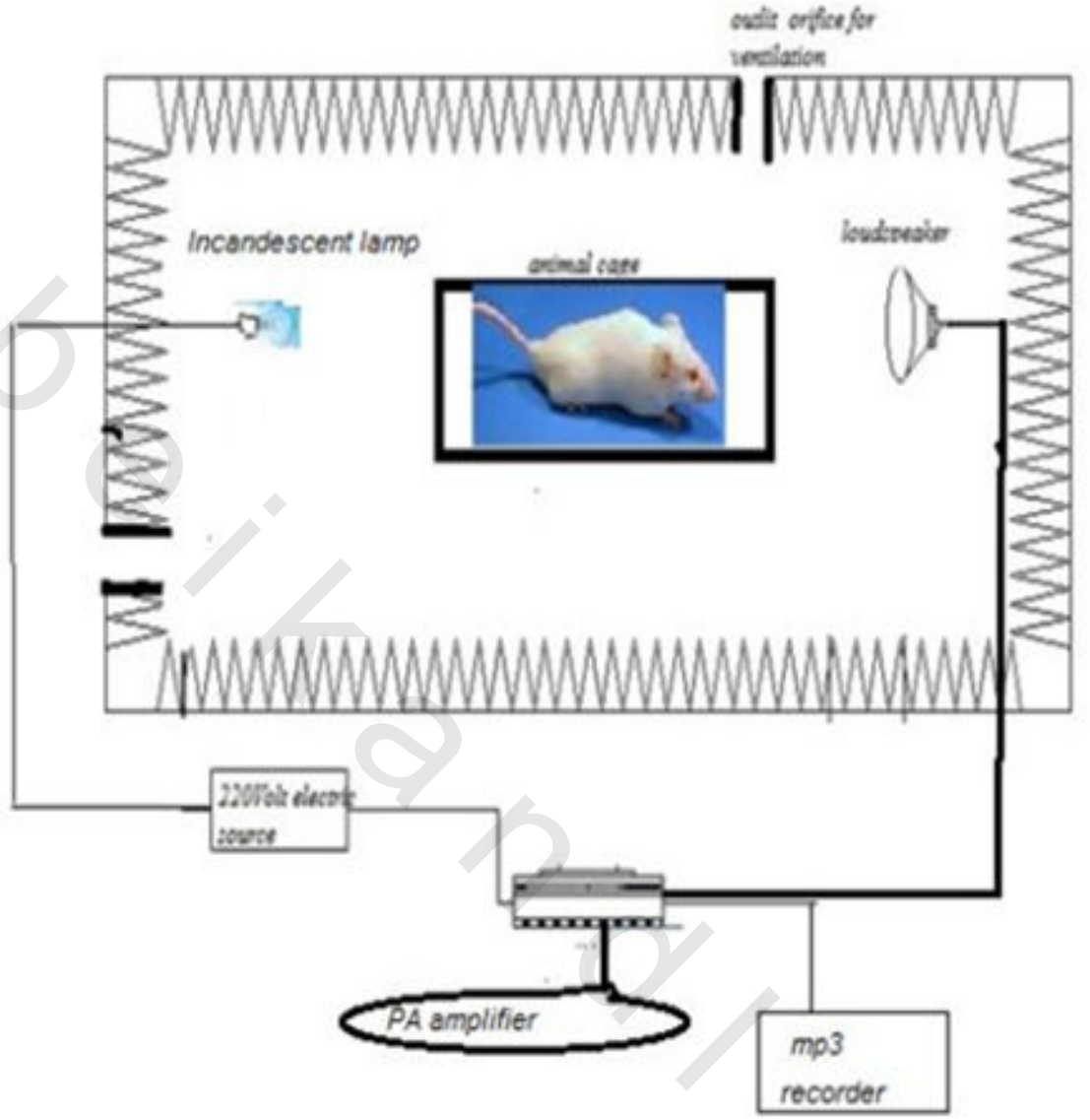
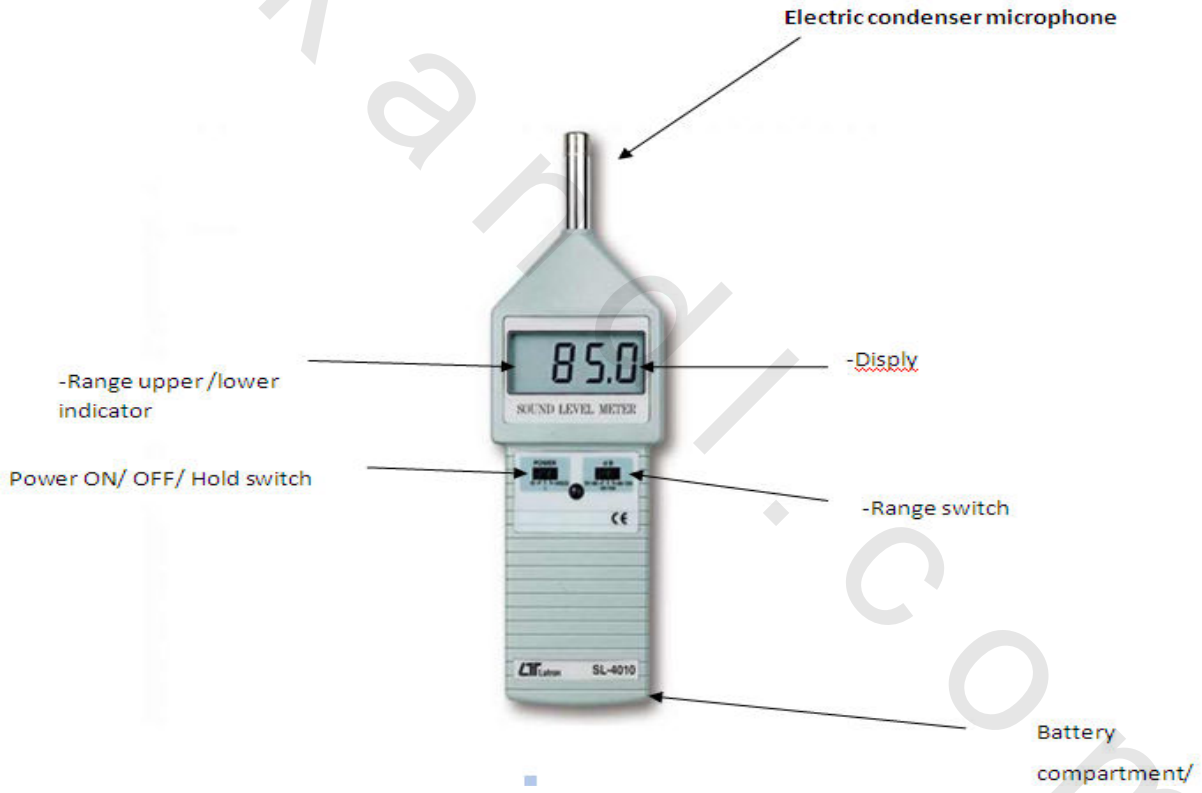


Fig.(15): Schematic diagram of the anechoic chamber used for mice exposure to the MRI sound .

4.3.Methods:

Recording the actual acoustic sound from MRI system closed type 1.5T (***Siemens AG 2005 Germany***)(see Fig.16) was recorded on to mp3, during examination at Nuor AlEslam Hospital for27-minute.The intensity of sound measured in decibels (dB) was measured and adjusted using audiometer[(***Lutron Make Digital Sound Level Meter***) ***India, Hyderabad, A.P***.] (see Fig.17). The actual sound level was ≈ 100 dB measured, the noisewasproduced bythree turbo loudspeaker (≈ 20-40 watt rms) driven by noise generator, and transmitted from speakers placed 10 cm away from the cage.

Fig.(16): MRI system closed type



Fig(17):Suppliers Digital Portable type Sound Level Meters

4.4.Sound analysis:

Sound spectrum analysis was doneaccording to the procedure mentioned by Chang et al (2012)[104].The recoded sound was examined using a laptop fitted with ***audacity soft-war*** and used ***True RTA soft-war known Real Time Audio***.

4.4.1.Plot spectrum:

Audacity software was used to record specified noise.Then we dealt with the selected audio (which is a set of sound pressure values at points in time) and converted it to a graph of frequencies (the horizontal scale in Hz) againstamplitudes (the vertical scale in dB).

4.5. Mice tissues samples:

At the end of the exposure period, mice were sacrificed without anesthesia. Blood samples werewithdrawn from all mice for the assessment of:

- Total and differential white blood cells (WBCs) count.[105]

- Blood glocuse level.[106]

Brain tissues were removed immediately, and devided into two portion, one portion for the dielectric measurements[107] and the other for biochemical investigation.

4.5.1.Preparation of crude homogenate for determination of total malonyldiadehyde (MDA)[108] , Superoxide dismutase (SOD)[109]and total protein:

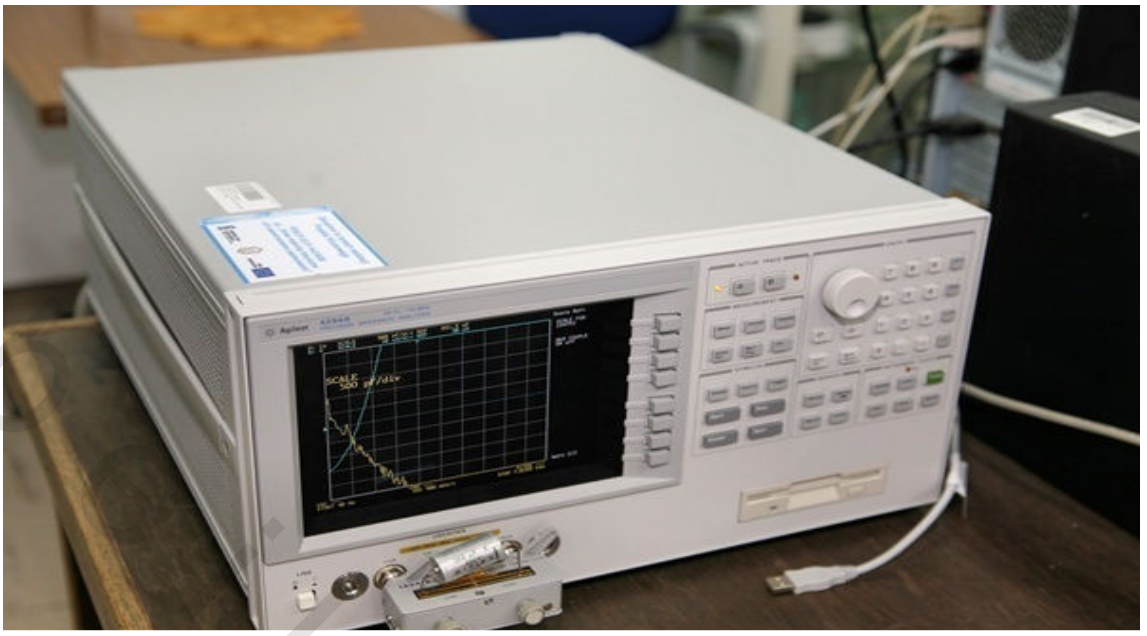
The 1 gm of the brain was homogenized in **5** volumes of cold phosphate buffer saline (0.1 M PBS,pH 7.4) using teflon-glass cold automatic Wheaton homogenizer. The final homogenate was then divided as follows:

- An aliquot of the totalhomogenate was used for thedetermination of malonyldiadehyde (MDA).
- An aliquot of the total homogenate was sonicated, then centrifuged at 3200xg for 20 minutes at 5C° and the supernatant was stored at -80 C° for assy ofSuperoxide dismutase.

4.6.BiophysicalMeasurement:

4.6.1.Dielectric Measurement:

Dielectric properties of the desired brain tissues from all groups were determined by RCL meter. Capacitance (C), resistance (R) and inductance (L) of tissues were carried out by using RCL meter. **Model FLUKE PM6306 PROGRAMMABLE AUTOMATIC RCL METER**, Fig.(18) the measured values were used to calculate the relative permittivity, conductivity and the dielectric loss. The RCL meter operated at frequency range from 100 KHz to 1000 KHz with accuracy of 0.05% the measured resultes were displayed on the high quality LCD monitor with decimal points and units test conditions can be stored and recalled from internal memory that will reduce setup for measurement preparing. Laboratory constructed parallels measuring capacitor cell was used.



Fig(18): RCL meter

4.6.2.Dielectric measurements:

In order to perform measurement on tissue cells using RCL Bridge an electric cell was designed for this propose. The tissues samples were connected to the RCL meter by means of two parallel silver electrodes coated with a silver chloride (Ag – Ag Cl electrodes). Silver – silver chloride provides agood contact transfer with minimum polarization.Tissue specimens were inserted in contact between the two electrodes as shown in Fig. (19).

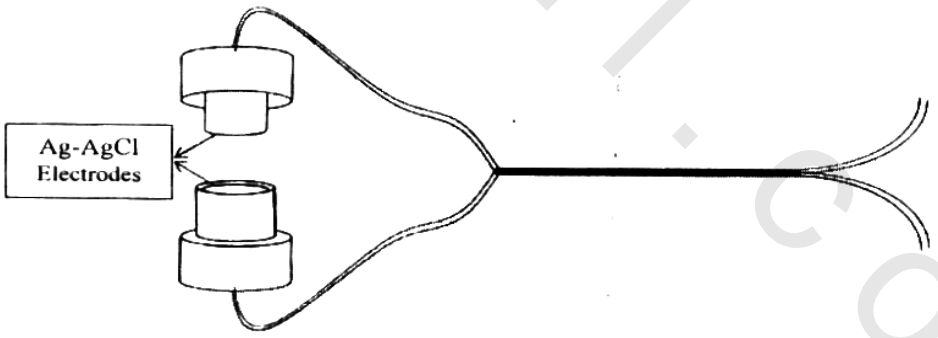


Fig.(19): A schematic diagram of the home made electric cell used for dielectric measurements.

The electrodes were held in position during measurements by means of gallows likes stands. The gallows stabs were used only for convenience in supporting the movable electrodes. The relative permittivity ε' and conductivity σ' were calculated from the equations [110]

$$C = \varepsilon' \varepsilon_0 A/d \qquad (15)$$

$G = \sigma' A / d$(16)

Where C(farad), G(Siemens), are the capacitance and conductance of the capacitor between the two measuring electrodes, A(m^2) is the surface area of the electrodes, d(m) the separation between the two electrodes, ε' the relative permittivity (Farad/meter), ε_0 the permittivity of vacuum ($8.85 * 10^{-12}$ F/m), and σ' the electrical conductivity(Siemens/m).

Calculated according to the relation.[111]

$$\varepsilon' = (\sigma' - \sigma_L) / 2\Pi f \varepsilon \quad (17)$$

$$\sigma' = 2\Pi f \varepsilon_0 (\varepsilon' - \varepsilon_h) \quad (18)$$

Where σ_L is low frequency limiting conductivity taken at 100 KHz, and ε_h is the high frequency limiting permittivity taken at 1000 KHz.

4.7. Biochemical Measurement:

4.7.1. Determination of Fasting blood glucose [112]

4.7.1.1.Principle:

Glucose is determined after enzymatic oxidation in the presence of glucose oxidase (GOD).The formed hydrogen peroxide reacts under catalysis of peroxidase with phenol and 4-aminoantipyrine to form a red violet quinoneimine dye as indicator.

$$\text{Glucose} + 2H_2O + O2 \xrightarrow{\text{GOD}} \text{Gluconic acid} + H_2O_2$$

$$2H_2O_2 + \text{Phenol} + \text{4-amino antipyrine} \xrightarrow{\text{peroxidase}} 4H_2O + \text{Quinoneim}$$

4.7.1.2. Reagents:

Phosphate Buffer	100 mmol/l
Phenol	4.0 mmol/l
4-amino-antipyrine	1.0 mmol/l
Glucose oxidase	>20 KU/l
Peroxidase	>2.0KU/l
Sodium Azide	8 mmol/l
Glucose standard(st)	100 mg/dl 5.55mmol/l

4.7.1.3.Procedure:

- 1 ml of enzyme reagent was mixed in test tubes with 10µl of plasma sample or glucose standard.

- The mixture was incubated at 37℃ for 10 minutes.
- Reagent blank was run through the same procedure.
- The absorbance of standard (A standard) and the sample(A sample) were measured against reagent blank at 546 nm within 30 minutes.

- **Calculation**

$$\text{- Glucose concentration (mg/dl)} = \frac{\text{A specimen}}{\text{A standard}} \times \text{standard concentration}$$

Where standard concentration was 100 mg/dl**.**

4.7. 2.Determination of superoxide dismutase(SOD) activity:

Superoxide dismutase(SOD)activity is determined by pyrogallol method of Marklund and Marklund.[113]

4.7.2.1. Principle:

The method depends on the spontaneous autoxidation of pyrogallol at alkaline pH, resulting in the production of superoxide anion radicals ($O\cdot 2^{-}$) which in turn enhance autoxidation of pyrogallol. Autoxidation is manifested as an increase in absorbance at 420 nm. The presence of SOD in the reaction leads to the removal of superoxide anion radical, thereby inhibiting autoxidation of pyrogallol.

This inhibition was linear up to 50% of the rate of reaction. A standard curve (Fig.20) was constructed by plotting the amount of SOD versus the corresponding percentage of inhibition of autoxidation.

The SOD in the sample was calculated from the standard curve and was expressed as mU/mg protein. One unit of SOD activity is defined as the amount of enzyme which inhibits the rate of autoxidation of pyrogallol by 50%.

4.7.2.2.Reagents:

- Tris-HCl buffer 50 mM containing 1 mMdiethylenetriaminepentacetic acid (DTPA), the pH of which was adjusted to 8.2 with 0.1 NHCl.
- Pyrogallol(Aldrich Chemical Co., Milwauke, Wisconsen, USA) 20 mM in 10 mM hydrochloric acid.
- A standard solution of SOD from horseradish(Sigma Chemical Co., Poole, England). was prepared in distilled water in a concentration of 5ng/µl.

4.7.2. 3. Procedure:

The reaction was carried out at 30°C in a quartz cuvette with a 1 cm light bath. The assay mixture contained 1ml of Tris-HCl buffer (containing 1mM DTPA) pH 8.2 and 30µl of the sample. The reaction started by addition of 10µl of pyrogallol. The change in the absorbance at 420 nm was determined through one minute (ΔA/min) using spectronic 21 spectrophoto-meter. The reference cuvette contained buffer instead of enzyme.

Standard curve (Fig.20) was made by the same procedure for the sample using different volumes of the standard SOD solution (5,10,15,20,25,30µl).

Calculation:

Percentage of inhibition of pyrogallol autoxidation =

$$100\text{-}\frac{\Delta A/\min(sample)}{\Delta A/\min(blank)}\times 100$$

The percentage inhibition of pyrogalol autoxidation by standard SOD was determined from the equation:

$$100\text{ -}\frac{\Delta A/\min(s\tan dard)}{\Delta A/\min(blank)}\times 100$$

From the standard curve, one unit of SOD activity equals 104 ng. The sample enzyme activity in mU/mg protein was obtained by dividing the value in ng/mg protein by 104 and multiplying it by 1000 to convert U to mU.

The specific activity of SOD in the sample as ng/mg protein was calculated by dividing the value of SOD as ng/ml by protein concentration in the sample (mg protein/ml).

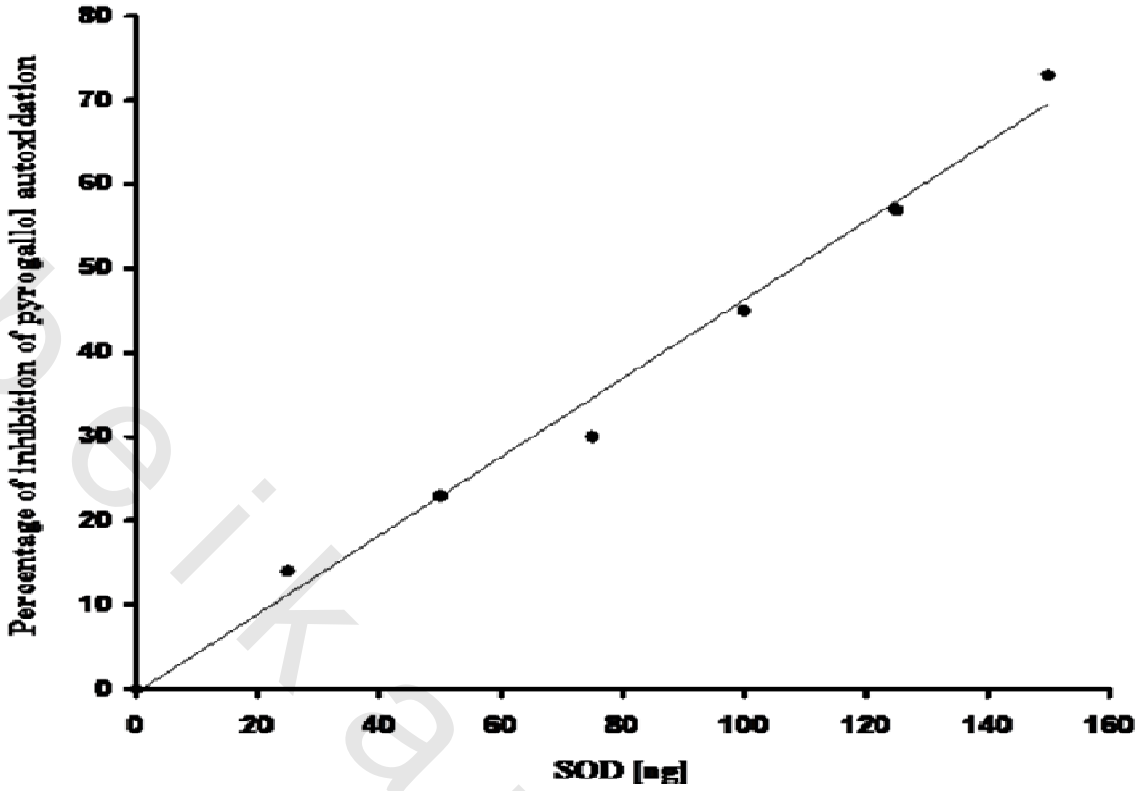


Figure (20): Standard curve of SOD.

4.7.3. Determination of Malondialdehyde (MDA):

Malondialdehyde in whole homogenate was determined according to the method of Draper and Hadley.[114]

4.7.3.1.Principle:

The sample under test is heated with thiobarbituric acid (TBA) at low pH. The resulting pink chromogen has a maximal absorbance at 532nm.

4.7.3.2.Reagents:

- Sodium dodecyl sulphate (SDS) 8.1 % in distilled water.
- Acetic acid 20%. Its pH was adjusted to 3.5 with 1N sodium hydroxide.
- Thiobarbituric acid (TBA) 0.8 % in distilled water.
- n-Butanol.

4.7.3.3.Procedure:

An aliquot of 0.1ml of the sample was pipette into a tube containing an equal volume of SDS solution. This was followed by the addition of 0.75ml acetic acid, 0.75ml of TBA and 0.3ml of distilled water. The contents of the tubes were then mixed with a vortex. The tubes were incubated in a boiling water bath for 1 hour then cooled to room temperature.

An aliquot of 0.5ml of distilled water was added to each tube followed by the addition of 2.5ml n-butanol. The contents of the tubes were vigorously mixed with a vortex then rotated in a centrifuge at 2500xg for 10 minutes. Absorbance of the organic layer was read at 532nm in a spectronic 21 spectrophotometer against a blank prepared and treated exactly like the sample however, containing phosphate buffer solution instead of the sample.

The concentration (nmole/ml) of MDA in sample was obtained from a standard curve (Figure.21) made by preparing serial dilutions of tetramethoxypropane (TMP), 1,2,4,6,8,12 nmole/ml, (Aldrich Chemical Co., Milwauke, Wisconsen, USA) in ethanol and treating them like the sample. Results were expressed as nmole MDA/mg protein by dividing the concentration of MDA in the sample by the protein concentration in the same sample.

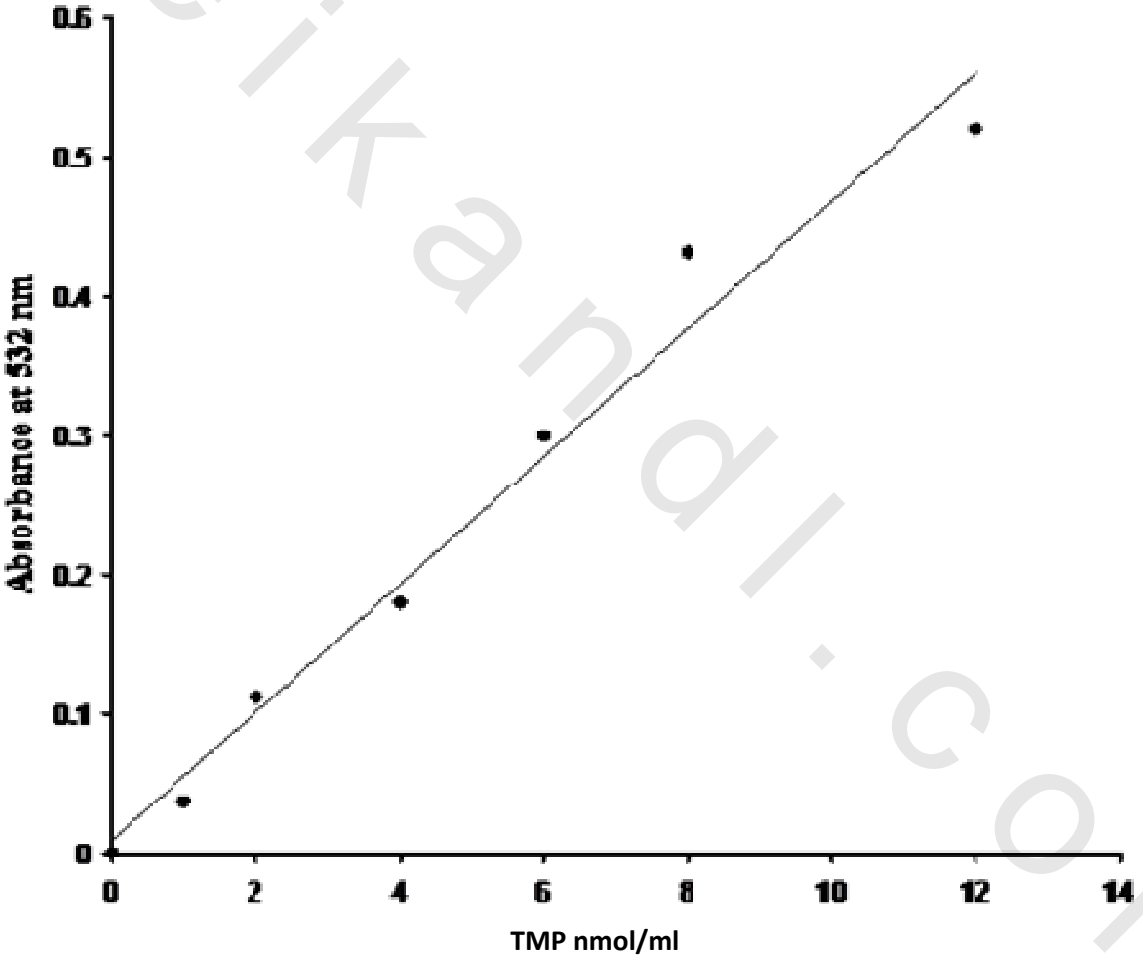


Fig.(21):Standard Curve of Malondialdehyde.

4.7.4.Determination of total proteins: [115]

4.7.4.1.Principle:

The method of Lowry et al. (1951)was used for the determination of the total protein in the samples.The colour produced is thought to be due to a complex between the alkaline copper-phenol reagent, tyrosine and tryptophan residues of the protein sample was estimated by referring to a standard curve Fig.(22)which was constructed using bovine serum albumin, fraction (V)(Sigma Chemical Co.,Poole,England).

4.7.4. 2. Reagent:

1. 0.1N sodium hydroxide.

2- 2% sodium carbonate (anhydrous)in 0.1 N NaOH.

3- 2g K/Na tartarate dissolved in 100 ml H_2O.

4- 1 g copper sulphate dissolved in 100 ml H_2O.

5- Lowry C reagent: prepared immediately before use by mixing volumes of sodium carbonate K/Na tartarate and copper sulphate reagents in a ratio of 100:1:1.

6- Folin-Ciocalteureagent(Sigma ChemicalCo.,Poole, England).The working reagent was prepared by diluting the stock reagent 1:1(v/v) with distilled water immediately before use.

4.7.4.3.Procedure:

- Aliquots of 10 µl of the diluted samples (1:10)weremixed with 2.5 ml of Lowry C-reagent.
- After incubation for 10 min at room temperature (in dark place),0.25 ml 0f working Folin- Ciocalteu reagent was added,mixed and incubated for 1 hour at room temperature, then the absorbance was read at 695 nm.
- In the blank dist. Water was added instead of the sample.
- The total protein concentrations in the samples were computed with,reference to the protein standard curve Fig.(22).

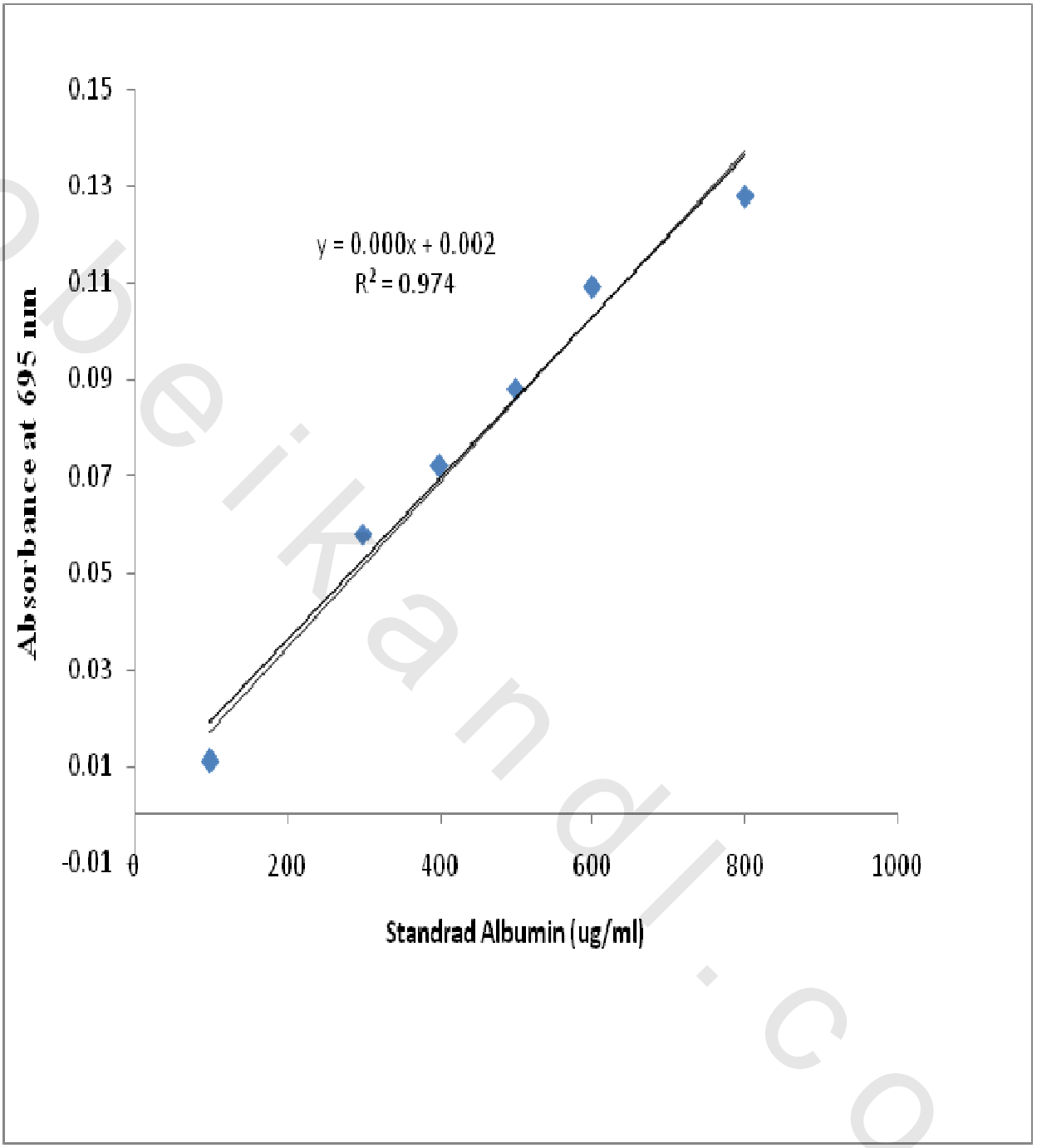


Fig.(22).The Standard Curve of Protein

4.8.Counting the total red blood cells: [116]

4. 8.1. Chemicals

Diluent:This is based on formal- citrate solution:

-1 % Formaline (10 ml/1. 40% formaldehyde) in 31.3 g/1. trisodium citrate.

4. 8.2. Methods:

A 1: 200 dilution of blood is made in formal- citrate solution. The cell suspention mixed by hand for 2 minutes combined with rotation. The counting chamber filled in one action and that no fluid flows into the surrounding moat. The cells counted using x 10 eye pieces. With a Neubauer chamber (figure. 23), the cells in four horizontal rectangles of 1 mm x 0.05 (80 small squares) counted including the cells which touch the top and right-hand margins and omitting from the count those which the bottom and left-hand margins of the small squares.

4.8.3.Calculation:

Number of cells in 80 small squares, 0.1mm in depth (0.02 mm^3 in volume) =N Red-cell count in millions per mm^3

=N x (1/0.02) x 200 (dilution) = N x 10,000

4.8.2. Platelets counts

4.8.2.1. Chemicals:

Diluent: This is based on formal- citrate red- cell dilutent:

- 1 % formaline (10 ml/1. 40% formaldehyde) in 31.3 g/1. Trisodium citrate.

4.8.2.2. Methods

A 1: 100 dilution of blood is made in formal- citrate solution by adding 0.02 ml of blood to 2 ml of dilutent. The cell suspension mixed by hand for 2 minutes combined with rotation. The counting chamber filled with the cell suspension. The cells counted using x 10 eyepieces. The number of platelets in one or more areas of 1 mm^2 counted.

Calculation:

Number of platelets counted in an area of $1mm^2$ (0.1 mm^3 in volume) =N the number of platelets per mm^3

=N x 10 x 100 (dilution)

= N x 1,000

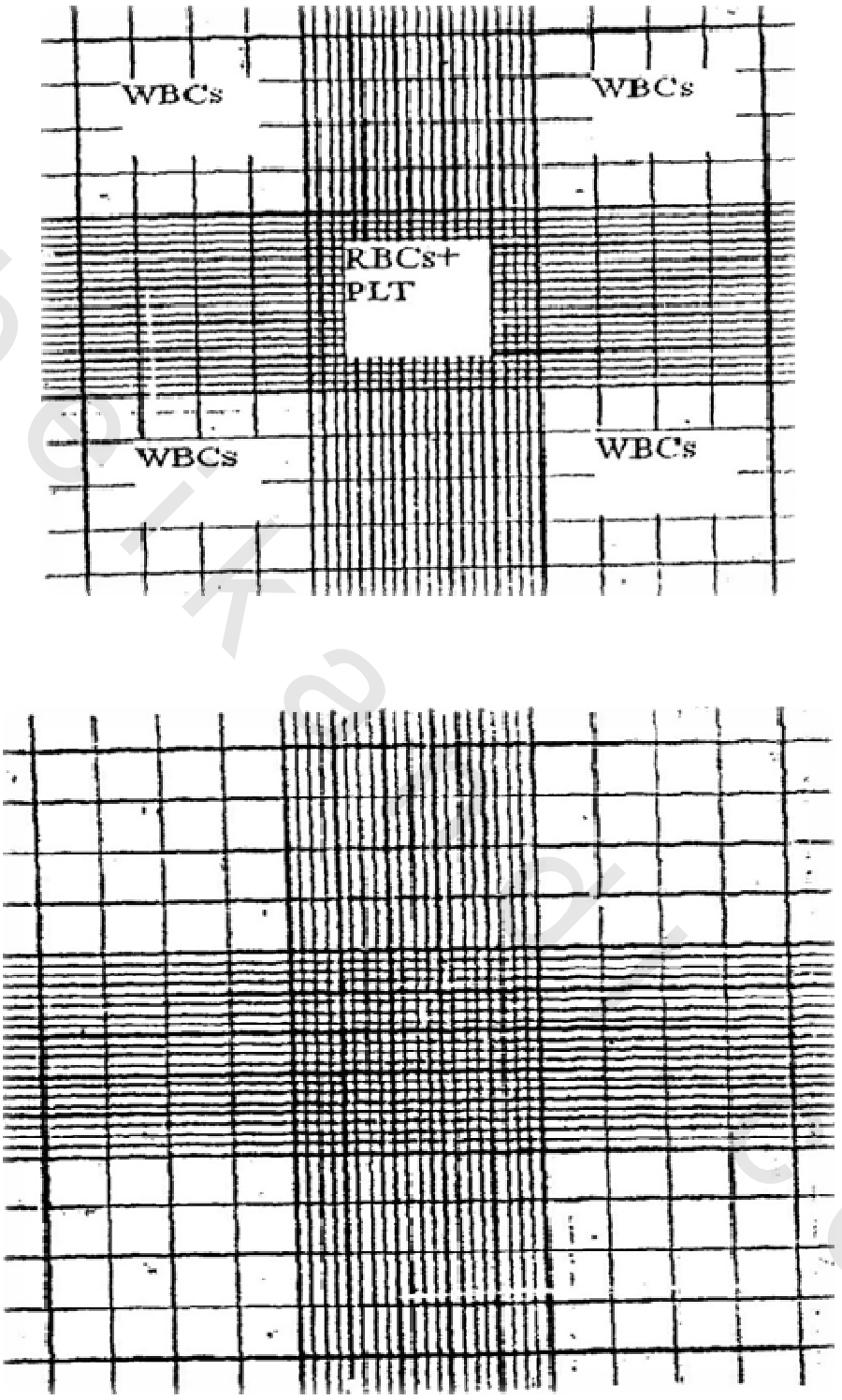


Figure (23): Improved Neubauer counting chamber. The central area consists of 25 groups of 16 small squares separated by closely ruled triple lines (whichappear as thick black lines in the figure).

4.8.3. Counting the total leucocytes blood cells

4.8.3.1. Chemicals:

Diluent: This is based on diluting fluid

- 2 % (20ml/1.) acetic aidcoloured pale violet with gentian violet**.**

4.8.3.2. Methods

A 1: 20 dilution of blood is made by adding 0.02 ml of blood to 0.38 ml of diluting fluid in a plastic tube. . The cell suspension mixed by hand for 2 minutes combined with rotation. The counting chamber filled by means of a Pasteur pipette. The cells counted using x 10 eyepieces. With a Neubauer chamber, he cells in four peripheral squares, counted including the cells which touch the top and right-hand margins and omitting from the count those which the bottom and left-hand margins of the small squares.

Calculation:

Number of cells in 4 peripheral squares,counted in 0.1 mm^3; N .The leucocytes count permm^3.

= N×10×20(dilution)

=N× 200

4.8.4. Differentialleucocytes count:

Differential leucocytes counts were performed by visual examination of blood films which were prepared on slides by the spread or wedge technique, which were executed as follow:

A small drop of blood was placed in the center line of a slide about I cm fromone end. Then, a spreader was placed in front of the drop at an angle of about 30° to the slide and moved it back to make contact with the drop.Thedrop spreaded out quickly along the line of contact. The spreader must not be lifted off until the last trace of blood has been spread out; with a correctly sized drop, the film should be about 3 cm in length. It is important that the film of blood finishes at least 1 cm before the end of the slide(Figure.24).

The film was left to dry in (the air and flood the slide with the Leishman's slain. After 2 min, double the volume of water was added and stained the film for 5-7 min. Then wash it in a stream of buffered water until it has acquired a pinkish tinge (up to2 min). After the back of the slide has been wiped clean, set it upright to dry. Different types of white blood cells were counted on blood films under light microscope (figure. 25).

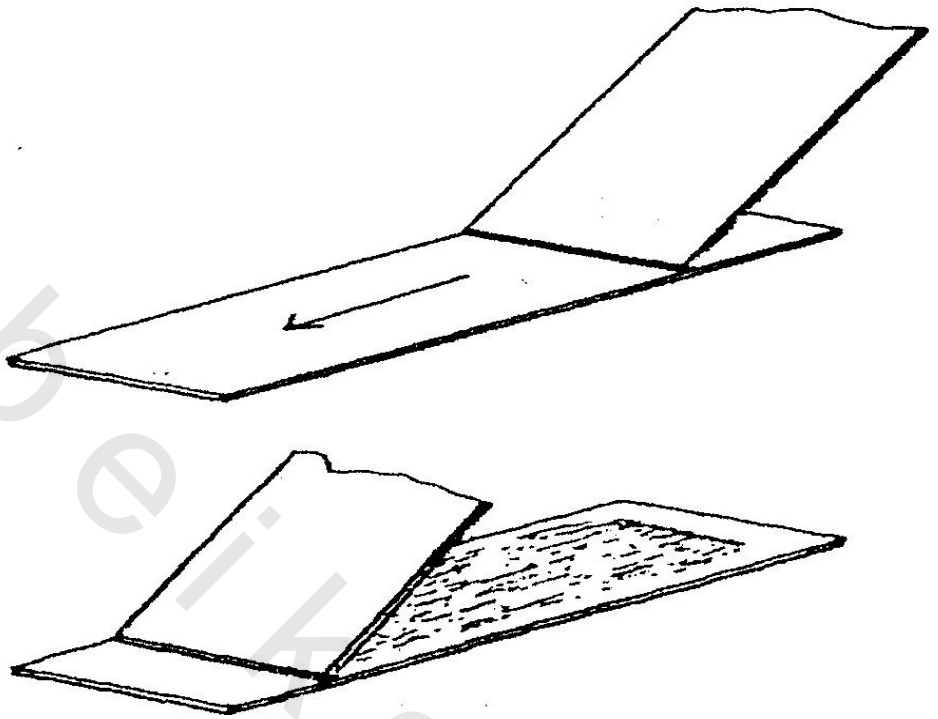


Figure (24): Slide method of preparation of blood smears.

Figure (25): Longitudinal method of counting cells in differential white cell counts.

RESULTS

The results will be presented as:

5.1- Sound analysis**.**

5.2- The effects acoustic noise of MRI system on the dielectric properties of brain tissues of different experimental animals groups.

5.3- The effects of acoustic noise of MRI system on the biochemical parameters of blood and brain tissues of different experimental animals groups.

RESULTS

5.1. Sound analysis:

Figure (26) shows the whole MRI recorded sound (using the ***audacity programmer***). Red color marks the selected band to be analyzed shown in Fig. (26). Cleary from this figure a homogenous sound and repeated bands. Fig (27) shows the expanded band of the selected portion of the recoded MRI noise. While the fully expanded MRI sound is shown in Fig (28).

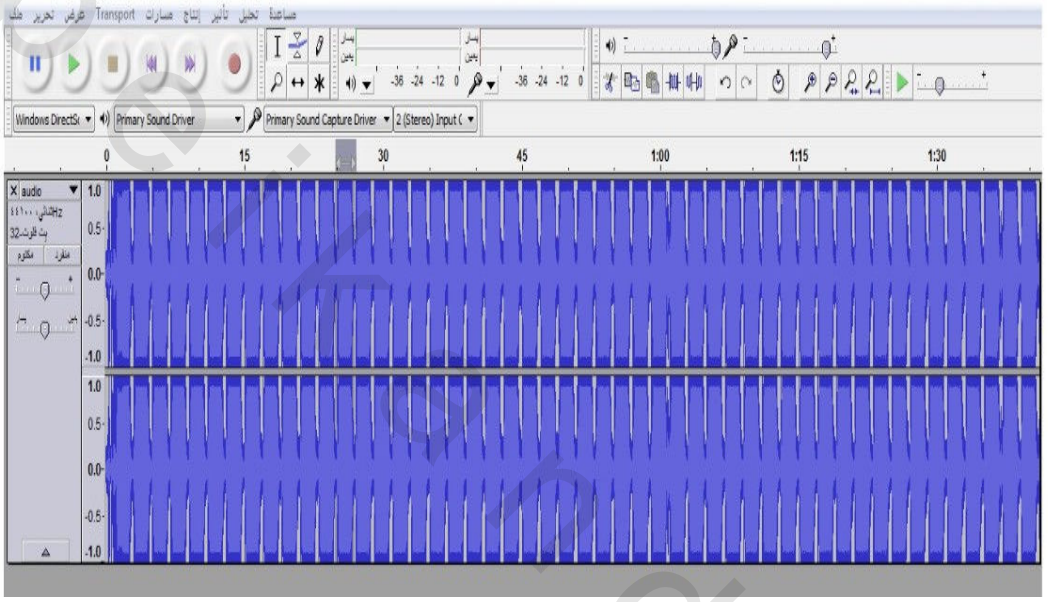


Fig.(26) :The MRI whole sound band in one.

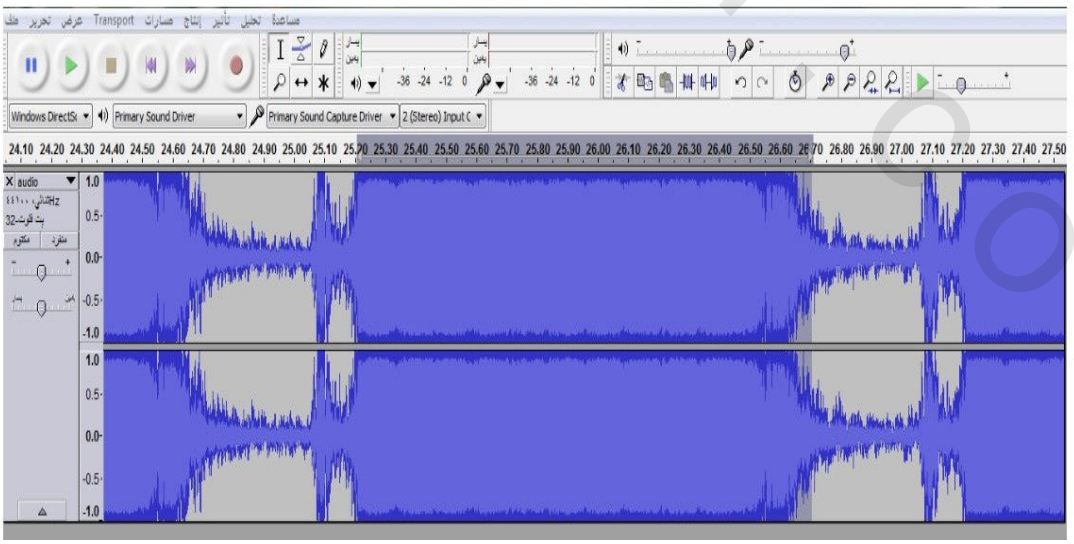


Fig (27): Expanded MRI sound.

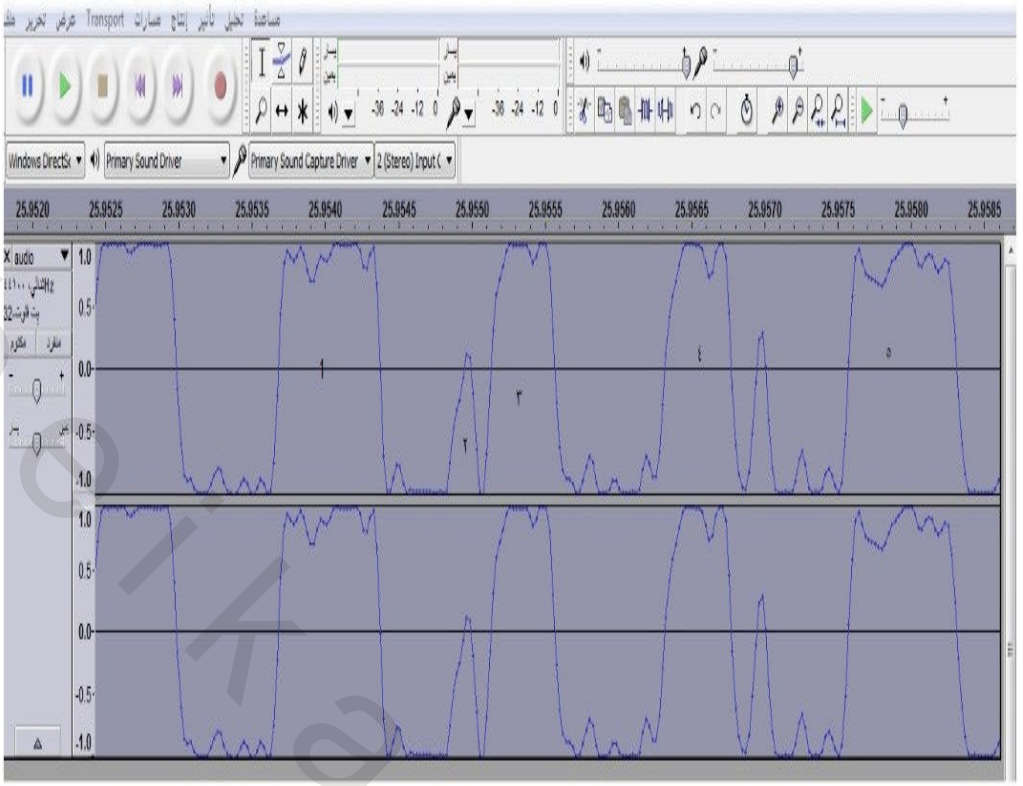


Fig.(28): Showing fully expanded MRI waveform bands labold 1 to 5. Bands 1,2 3, and 4 are the characteristic sound of the used MRI.

5.1.1. Predominant frequency for MRI sound:

The predominant frequency was determined using the unexpanded plot shown in Fig (26), which contains the entire recorded MRI signal sound. The ***Real Time Analyzer*** (see the ***audacity programmer***) was used in this analysis, to plot the frequancy-intensity diagram as seen in Fig(29). Clearly shows the distribution of frequancy-intensity bands the X-axis shows the frequency in KHz, the Y-axis shows the sound intensity values in dB. From this Figure the predominant frequency is the 1KHz , which a sound intensity of 105 dB.

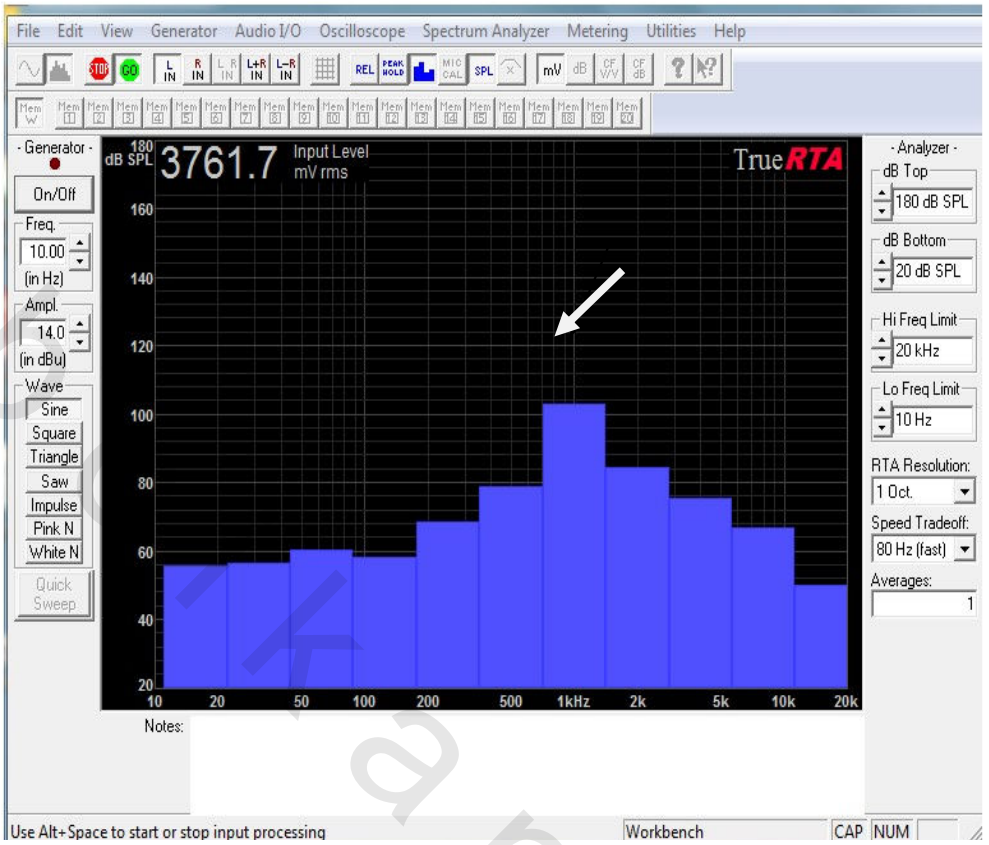


Fig.(29): Shows the MRI sound frequency-intensity distribution diagram (the arrow points out the predominant frequency in the used sound band).

5.2. Effect of MRI acoustic noise exposure on the dielectric measurements of brain tissues:

In the present work dielectric measurements were performed on brain of all studies groups (control and exposed to different modalities of noise). The uncertainty in the measured data was determined as standard deviation and found to be less than 1%. Scanning the frequency of the applied frequency from 100 kHz up to 1 MHz was performed while recording the variation of both capacitance (C) and resistance (R). The conductance was calculated as a reciprocal of R. These values were used to calculate the relative permittivity (ε') and conductivity (σ') of the brain tissues.

Conductivity and permittivity diagrams of brain tissue taken from control and exposed groups are shown in Figures (30,31). As seen in these diagrams the degree of depressed center and the maximum values of the real conductivity and permittivity depend on the noise intensities.

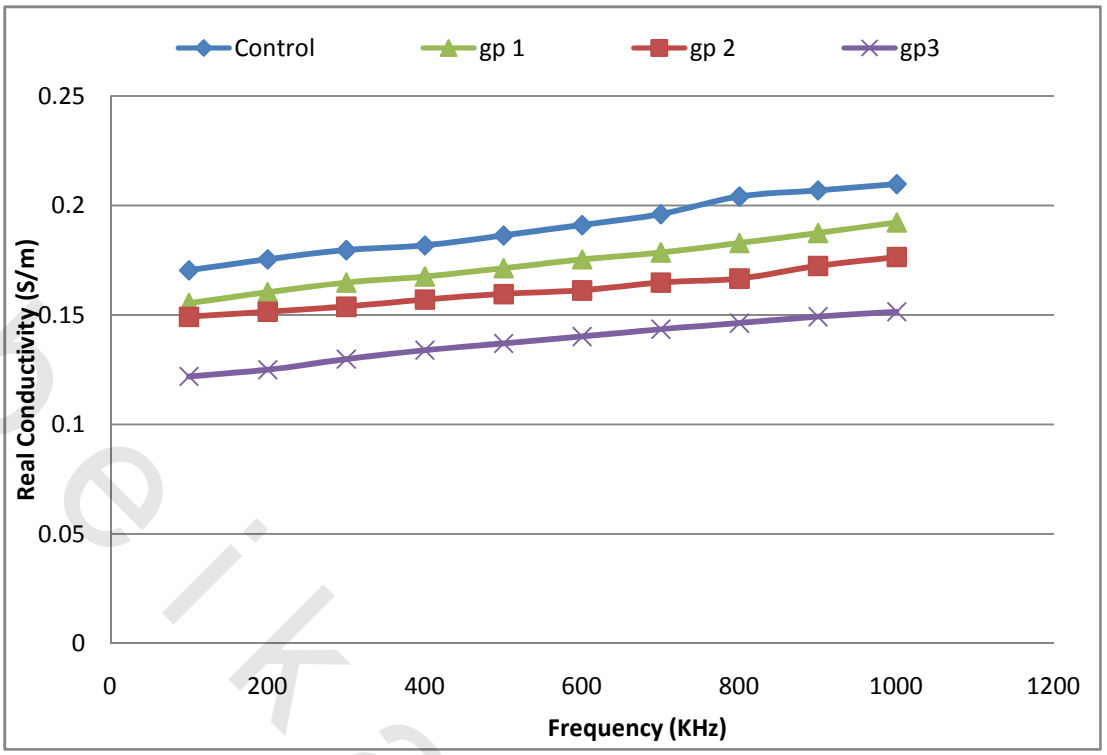


Fig (30): Variation of real conductivity with frequency of mice brain exposed to different MRI noise intensities.

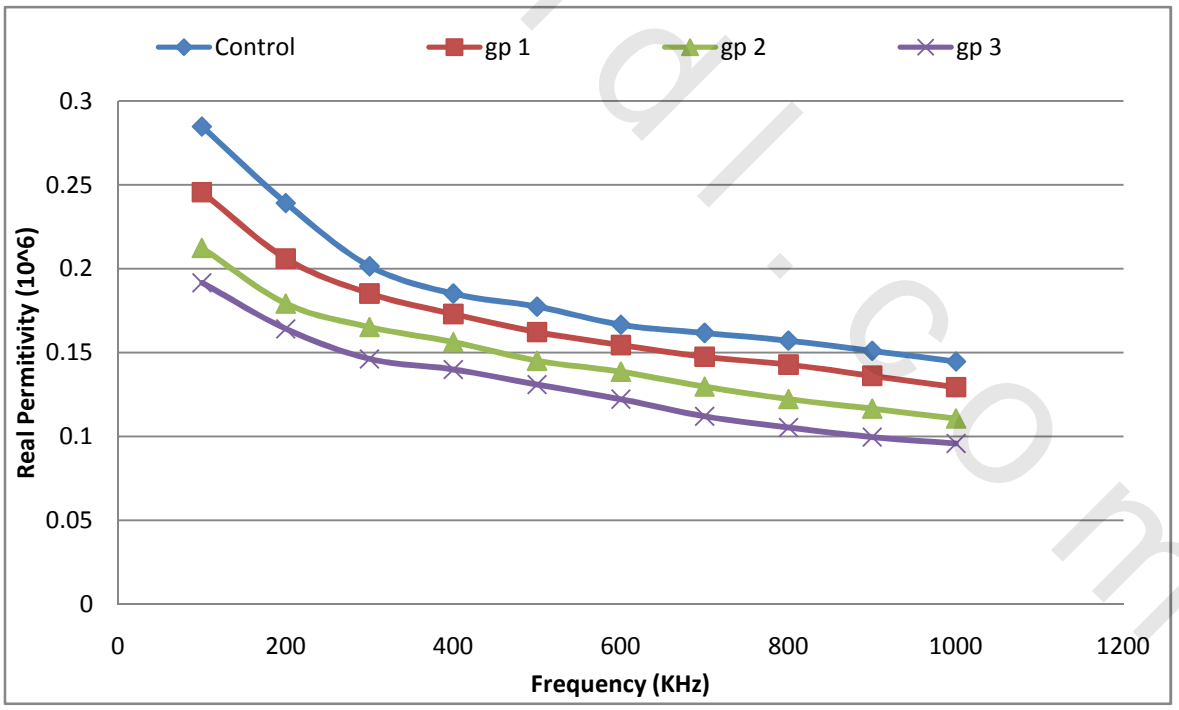


Fig (31): Variation of relative permittivity with frequency of mice brain exposed to different MRI noise intensities.

5.3. Effect of MRI acoustic noise exposure on some hematological and some biochemical parameters:

5.3.1. Effect of noise on blood glucose:

The individual data and the mean values ± S.D of blood glucose in all different studied groups are shown in Table (2). The statistical analysis of these results represented in Table (3) and Fig. (32).

It was demonstrated that blood glucose level ranged from 123-145 mg/dl with mean value of 128.4 ± 6.5 mg/dl in control group. In mice exposed to 121 dB (GB1) the blood glucose level ranged from 153.2-190 with mean value of 173.4 ± 12.6 mg/dl. In GB2 where mice exposed to 100 dB, the blood glucose level ranged from 160-190 with mean value of 171.6 ± 11.61 mg/dl. On the other hand, mice exposed to acoustic noise of 90 dB showed elevation in the blood glucose level ranged from 133-162.2 with mean of 148.1 ± 11.66 mg/dl.

Statistical analysis of blood glucose level (mg/dl) showed significant differences in the mean value in all exposed groups (GB1, GB2 and GB3) as compared to control groups ($P < 0.001$).

Table (2): Blood glucose levels (as mg/dl) of mice to studied experimental groups. (GB1 exposed to 121 dB, GB2 exposed to 100 dB, and GB3 exposed to 90 dB) in comparison with control group.

No.	Control	GB1	GB2	GB3
1	132	174	160.2	133
2	126	167	170.1	140.4
3	123	180	186	162.2
4	145	190	180.2	134
5	124	180	160	152.4
6	130	190	160	152
7	127	153.2	190	160
8	128	170.6	160.3	150.2
9	126	174	170.1	135.2
10	123	153.2	180	162
Min.	123.0	153.20	160.0	133.0
Max.	145.0	190.0	190.0	162.20
Mean	128.40	173.20	171.69	148.14
SD.	6.52	12.92	11.61	11.66

Table (3): Statistical analysis of Blood glucose levels (as mg/dl) all different studied groups.

	Control (I) (n=10)	**GB1 (II) (n=10)**	**GB2 (III) (n=10)**	**GB3 (IV) (n=10)**
Blood glucose levels (as mg/dl)				
Min. - Max.	123.0 – 145.0	153.20 – 190.0	160.0 – 190.0	133.0 – 162.20
Mean ± SD.	128.40 ± 6.52	173.20 ± 12.92	171.69 ± 11.61	148.14 ± 11.66
F	34.793*			
P	<0.001*			
p_1		<0.001*	<0.001*	0.005*
p_2			0.993	<0.001*
p_3				0.001*
Sig. bet. Grps	I-II*, I-III*, I-IV*, II-IV*			

F: F test (ANOVA)
p_1: p value for Post Hoc test (Scheffe) for comparing between control and each other group
p_2: p value for Post Hoc test (Scheffe) for comparing between GB1 with each of GB2 and GB3
p_3: p value for Post Hoc test (Scheffe) for comparing between GB2 and GB3
Sig. bet. grps was done using Post Hoc test (Scheffe)
*: Statistically significant at $p \leq 0.05$

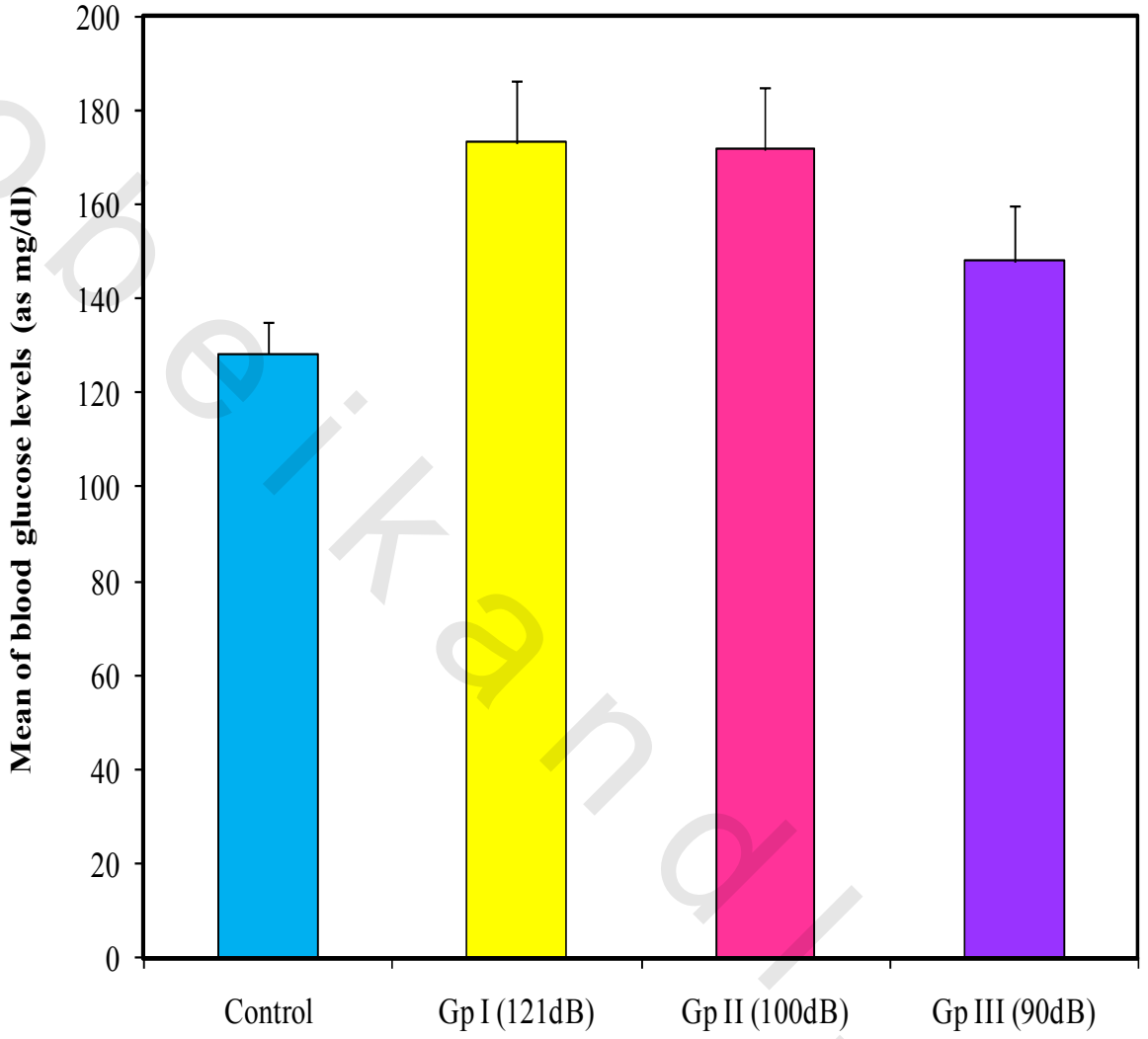


Figure (32): Effect of MRI noise exposure on Blood glucose levels (as mg/dl) all different studied groups.

5.3.2. Hematological analysis of mice exposed to MRI noise:

Standard hematology markers were selected for analysis such as hemoglobin level (Hb), hematocrit (HCT), red blood cells (RBCs), platelets (PLT), and white blood cells count (WBCs) due to exposure to different intensities of MRI noise. Samples were analyzed according to the method mentioned in pages (39-42). Obviously Hb, Ht, RBCs, MCV, MCH and MCHC were significantly decreased in all experimental groups as compared with control group. While WBCs and PLT were significantly increase in all experimental groups as compared the control group.

5.3.3. Hemoglobin levels:

Result changes in the hematological indices revealed significantly decreased in hemoglobin levels in all exposed groups as compared to control group ($p \leq 0.01$) . The statistical analysis of hemoglobin levels in all studied groups are shown in Table (4) and Fig.(33).

Table (4): Statistical analysis of Hemoglobin Hb (g/dl) levels all different studied groups.

	Control (I) (n=10)	**GB1 (II) (n=10)**	**GB2 (III) (n=10)**	**GB3 (IV) (n=10)**
Hb (g/dl)				
Min. - Max.	10.60 – 13.80	4.30 – 13.0	6.0 – 10.90	9.80 ± 15.0
Mean ± SD.	12.42 ± 0.98	9.89 ± 2.76	9.21 ± 1.89	12.05 ± 1.61
% change from control		↓20.37	↓25.85	↓2.98
F	6.784*			
P	0.001*			
p_1		0.049*	0.008**	0.980
p_2			0.890	0.116
p_3				0.022*
Sig. bet. grps	I-II*,I-III**,III-IV*			

F: F test (ANOVA)
p_1: p value for Post Hoc test (Scheffe) for comparing between control and each other group
p_2: p value for Post Hoc test (Scheffe) for comparing between GB1 with each of GB2 and GB3
p_3: p value for Post Hoc test (Scheffe) for comparing between GB2 and GB3
Sig. bet. grps was done using Post Hoc test (Scheffe)
*: Statistically significant at $p \leq 0.05$
**: Statistically significant at $p \leq 0.01$

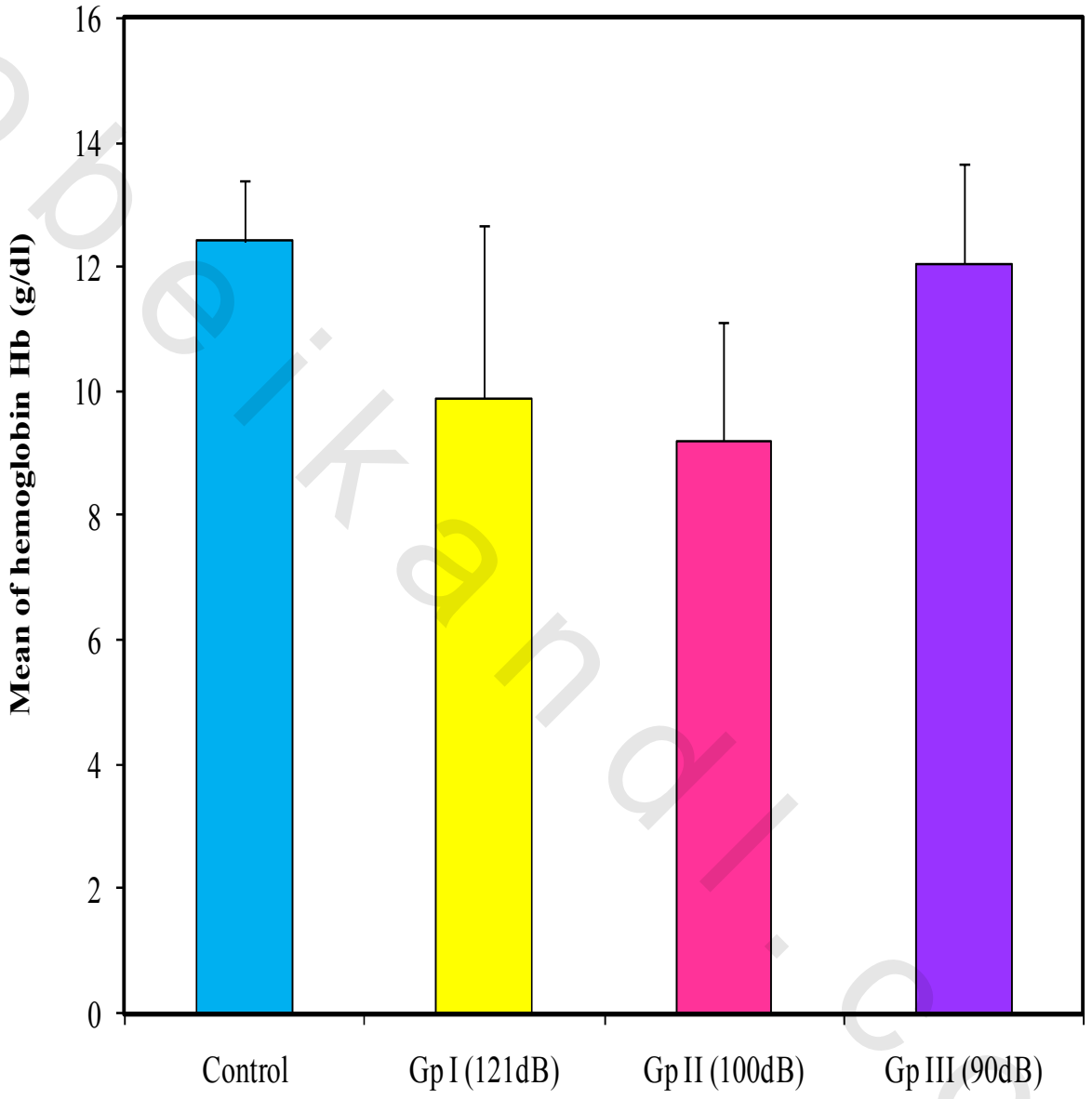


Figure (33): Effect of MRI noise exposure on hemoglobin levels.

5.3.4. Hematocrit levels:

Hematocrit percentage in experimental groups showed significant reduction as compared to the control group ($p \leq 0.018$).The statistical analysis of hematocrit are shown in Table (5) and Fig (34).

Table (5): Statistical analysis of Hematocrit HT (%) levels all different studied groups.

	Control (I) (n=10)	GB1 (II) (n=10)	GB2 (III) (n=10)	GB3 (IV) (n=10)
HT (%)				
Min. - Max.	45.90 – 32.60	14.60 – 45.0	22.50 – 37.60	32.40 – 44.90
Mean ± SD.	40.09 ± 4.69	32.80 ± 9.51	33.20 ± 4.94	39.06 ± 4.16
% change from control		↓18.18	↓17.19	↓2.57
F	3.791*			
P	0.018*			
p_1		0.094	0.124	0.987
p_2			0.999	0.186
p_3				0.235
Sig. bet. Grps	NS			

F: F test (ANOVA)
p_1: p value for Post Hoc test (Scheffe) for comparing between control and each other group
p_2: p value for Post Hoc test (Scheffe) for comparing between GB1 with each of GB2 and GB3
p_3: p value for Post Hoc test (Scheffe) for comparing between GB2 and GB3
Sig. bet. grps was done using Post Hoc test (Scheffe)
*: Statistically significant at $p \leq 0.05$

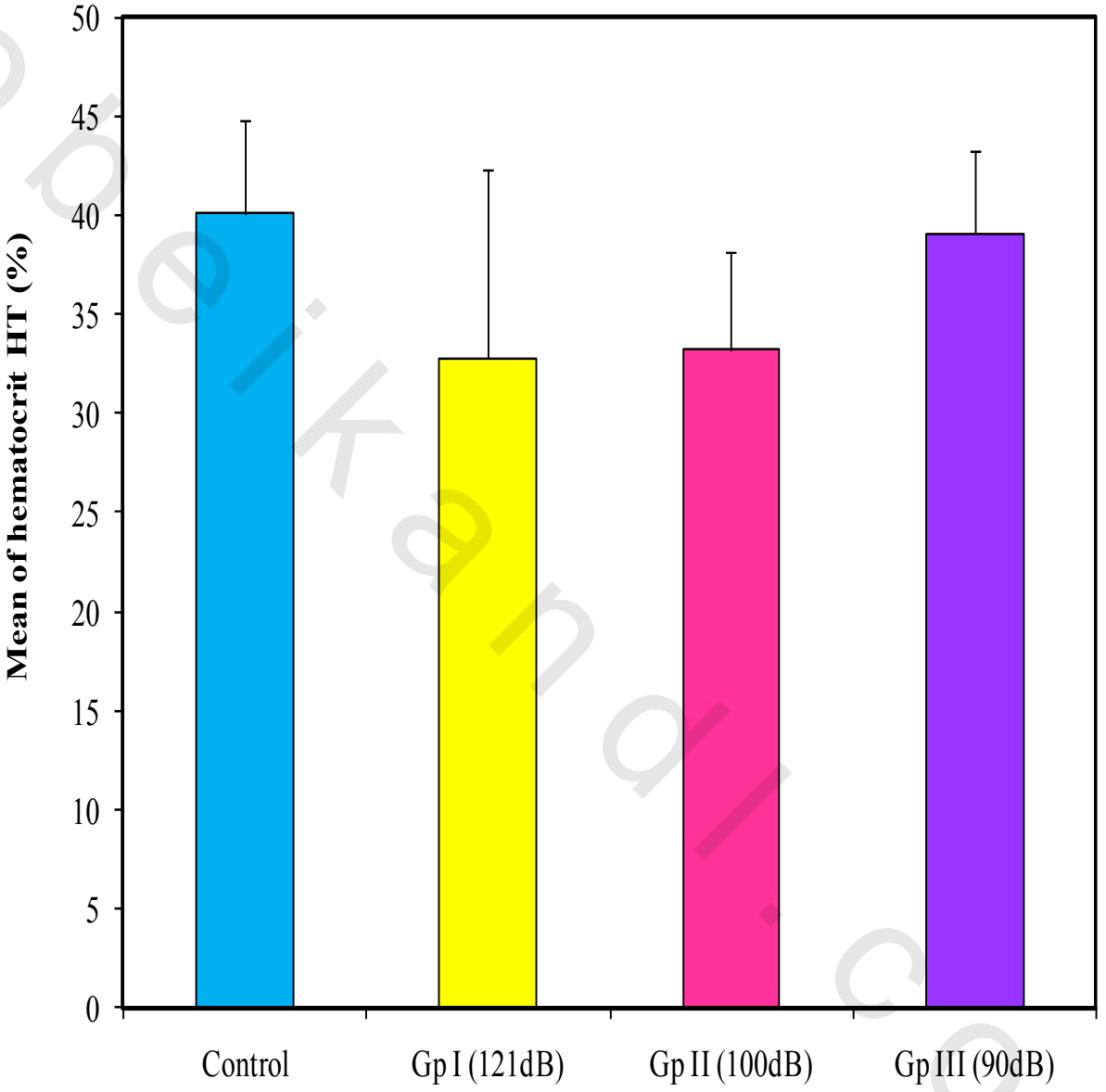


Figure (34): Effect of MRI noise exposure on hematocrit level.

5.3.5 Mean corpuscular volume (MCV):

MCV were significantly decreased for groups exposed the acute noise as compared to control ($p < 0.064$) .The statistical analysis of Mean corpuscular volume are shown in Table (6) and Fig.(35).

Table (6): Statistical analysis of Mean corpuscular volume (MCV) of all different studied groups.

	Control (I) (n=10)	GB1 (II) (n=10)	GB2 (III) (n=10)	GB3 (IV) (n=10)
MCV				
Min. - Max.	45.20 – 62.70	45.80 – 52.90	44.20 – 52.10	47.10 – 54.40
Mean ± SD.	51.34 ± 4.68	49.42 ± 2.44	47.72 ± 2.23	48.42 ± 2.16
% change from control		↓3.74	↓3.74	↓5.69
F	2.641			
P	0.064			
p_1		0.585	0.091	0.227
p_2			0.675	0.911
p_3				0.967
Sig. bet. grps	NS			

F: F test (ANOVA)
p_1: p value for Post Hoc test (Scheffe) for comparing between control and each other group
p_2: p value for Post Hoc test (Scheffe) for comparing between GB1 with each of GB2 and GB3
p_3: p value for Post Hoc test (Scheffe) for comparing between GB2 and GB3
Sig. bet. grps was done using Post Hoc test (Scheffe)

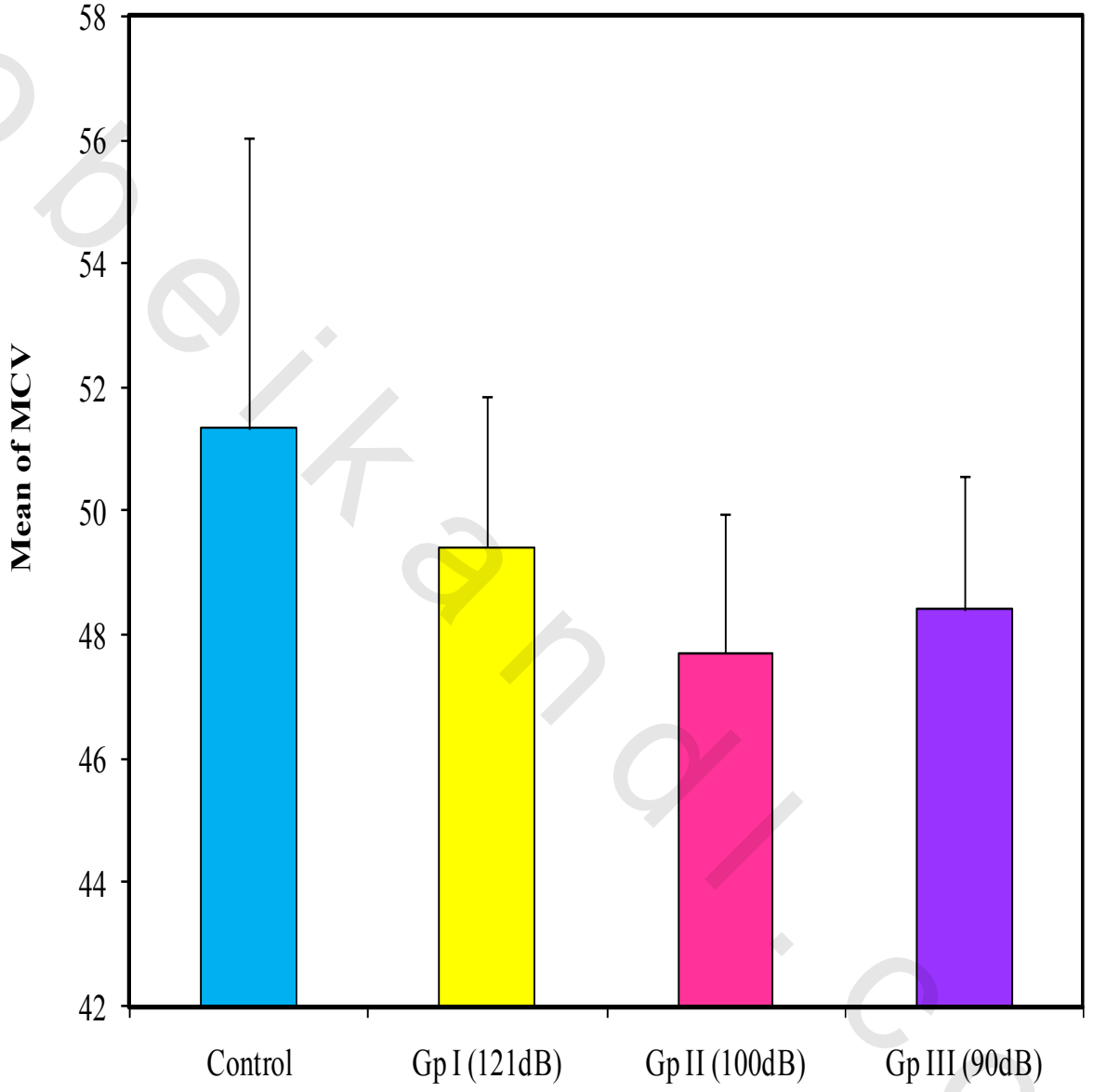


Figure (35): Effect of MRI noise exposure on Mean corpuscular volume (MCV).

5.3.6. Mean corpuscular hemoglobin (MCH):

MCH were significantly decreased for groups exposed the acute noise as compared to control ($p < 0.009$). The statistical analysis of Mean corpuscular hemoglobin are shown in Table (7) and Fig.(36).

Table (7): Statistical analysis of Mean corpuscular hemoglobin (MCH) of all different studied groups.

	Control (I) (n=10)	GB1 (II) (n=10)	GB2 (III) (n=10)	GB3 (IV) (n=10)
MCH				
Min. - Max.	14.30 – 16.90	13.50 – 16.20	7.20 – 15.80	12.90 – 16.20
Mean ± SD.	15.72 ± 0.74	14.94 ± 1.0	13.44 ± 2.33	14.59 ± 1.02
% change from control		↓4.96	↓14.50	↓7.19
F	4.484*			
P	0.009*			
p_1		0.680	0.011*	0.377
p_2			0.152	0.958
p_3				0.362
Sig. bet. grps	I-II*			

F: F test (ANOVA)
p_1: p value for Post Hoc test (Scheffe) for comparing between control and each other group
p_2: p value for Post Hoc test (Scheffe) for comparing between GB1 with each of GB2 and GB3
p_3: p value for Post Hoc test (Scheffe) for comparing between GB2 and GB3
Sig. bet. grps was done using Post Hoc test (Scheffe)
*: Statistically significant at $p \leq 0.05$

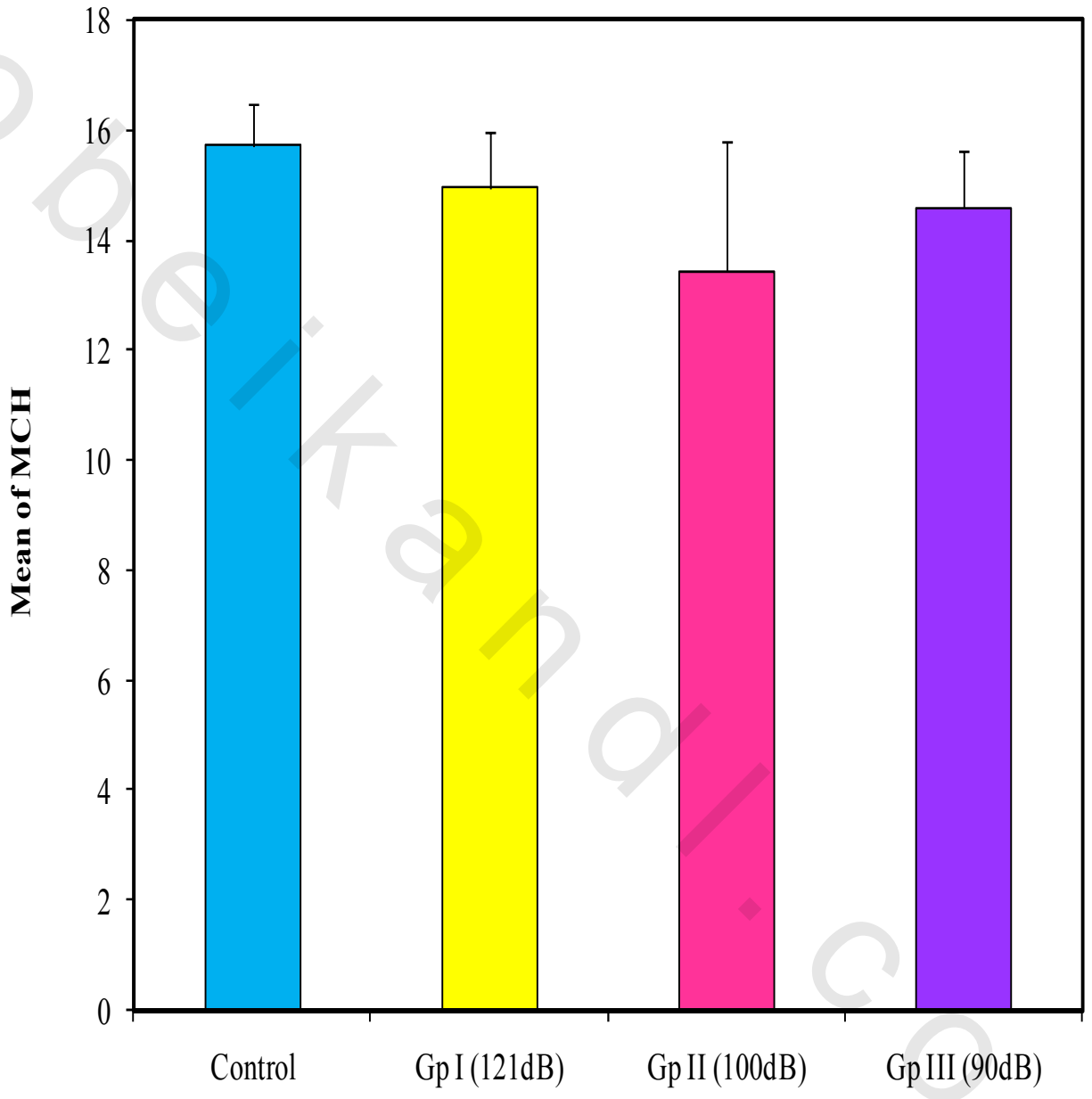


Figure (36): Effect of MRI noise exposure on Mean corpuscular hemoglobin (MCH).

5.3.7. Mean corpuscular hemoglobin concentration (MCHC):

MCHC were significantly decreased for groups exposed the acute noise as compared to control ($p < 0.125$). The statistical analysis of Mean corpuscular hemoglobin concentration shown in Table (8) and Fig.(37).

Table (8): Statistical analysis of Mean corpuscular hemoglobin concentration (MCHC) of all different studied groups.

	Control (I) (n=10)	GB1 (II) (n=10)	GB2 (III) (n=10)	GB3 (IV) (n=10)
MCHC				
Min. - Max.	28.70 – 32.80	28.90 – 32.50	28.0 – 30.80	26.10 – 31.50
Mean ± SD.	31.09 ± 1.71	30.22 ± 1.01	29.4± 0.84	28.95 ± 1.73
% change from control		↓2.80	↓12.87	↓6.88
F	2.046			
P	0.125			
p_1		0.968	0.163	0.673
p_2			0.359	0.908
p_3				0.760
Sig. bet. grps	NS			

F: F test (ANOVA)
p_1: p value for Post Hoc test (Scheffe) for comparing between control and each other group
p_2: p value for Post Hoc test (Scheffe) for comparing between GB1 with each of GB2 and GB3
p_3: p value for Post Hoc test (Scheffe) for comparing between GB2 and GB3
Sig. bet. grps was done using Post Hoc test (Scheffe)

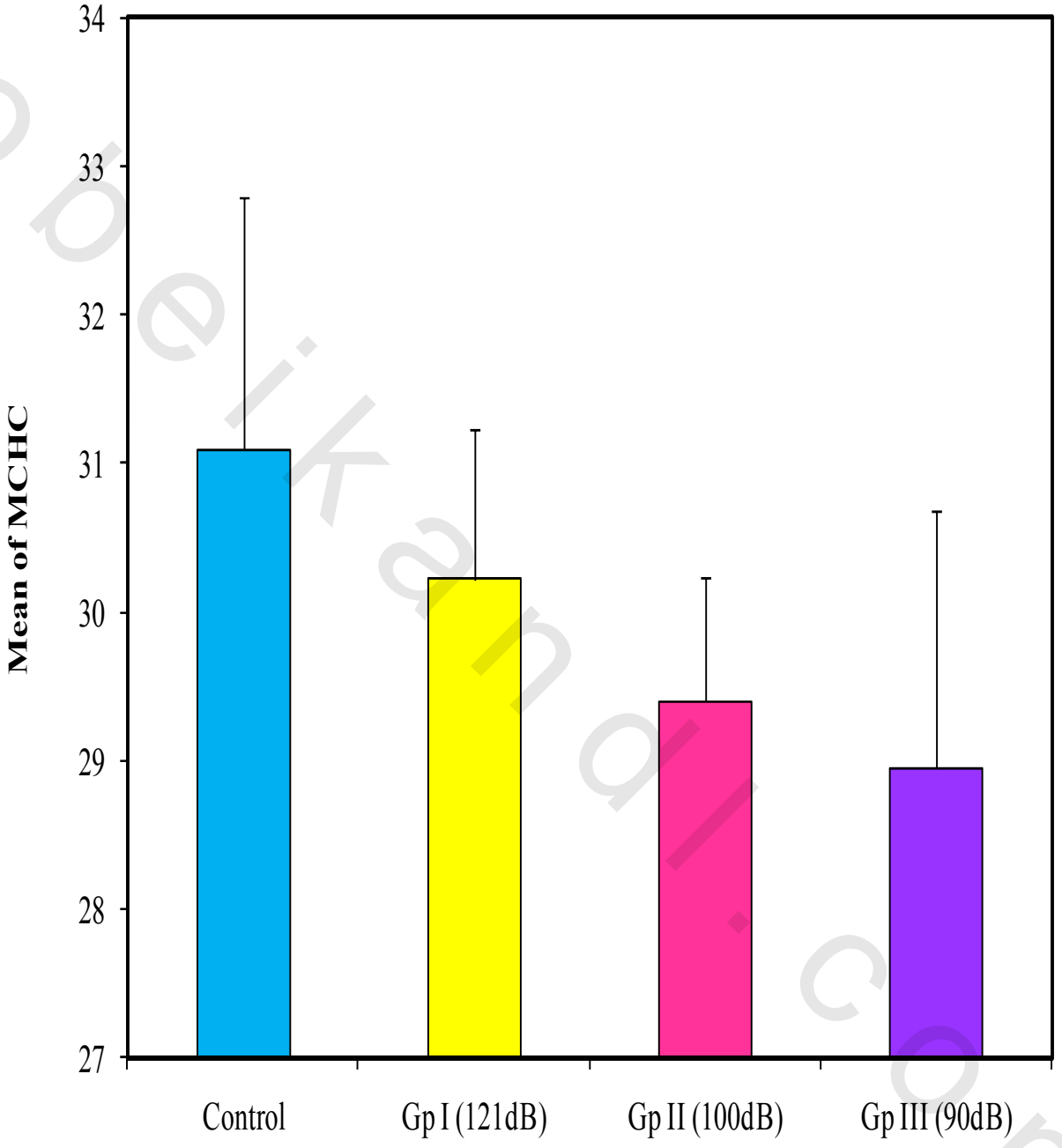


Figure (37): Effect of MRI noise exposure on Mean corpuscular hemoglobin concentration (MCHC).

5.3.8. Red blood cells counts:

Red blood cells counts were significantly decreased for all groups exposed the acute noise as compared to control ($p < 0.008$).The statistical analysis of red blood cells counts are shown in Table (9) and Fig.(38).

Table (9): Statistical analysis of red blood cells counts (RBC) ($\times 10^6$/µl) levels all different studied groups.

	Control (I) (n=10)	GB1 (II) (n=10)	GB2 (III) (n=10)	GB3 (IV) (n=10)
RBC ($\times 10^6$/µl)				
Min. - Max.	6.27 – 9.80	3.19 – 9.54	5.02 – 7.79	6.87 – 10.90
Mean ± SD.	8.33 ± 1.20	6.62 ± 1.88	6.81 ± 1.01	8.27 ± 1.19
% change from control		↓20.53	↓18.25	↓0.72
F	4.564*			
P	0.008*			
p_1		0.056	0.119	1.000
p_2			0.992	0.079
p_3				0.143
Sig. bet. grps	NS			

F: F test (ANOVA)
p_1: p value for Post Hoc test (Scheffe) for comparing between control and each other group
p_2: p value for Post Hoc test (Scheffe) for comparing between GB1 with each of GB2 and GB3
p_3: p value for Post Hoc test (Scheffe) for comparing between GB2 and GB3
Sig. bet. grps was done using Post Hoc test (Scheffe)
*: Statistically significant at $p \leq 0.05$

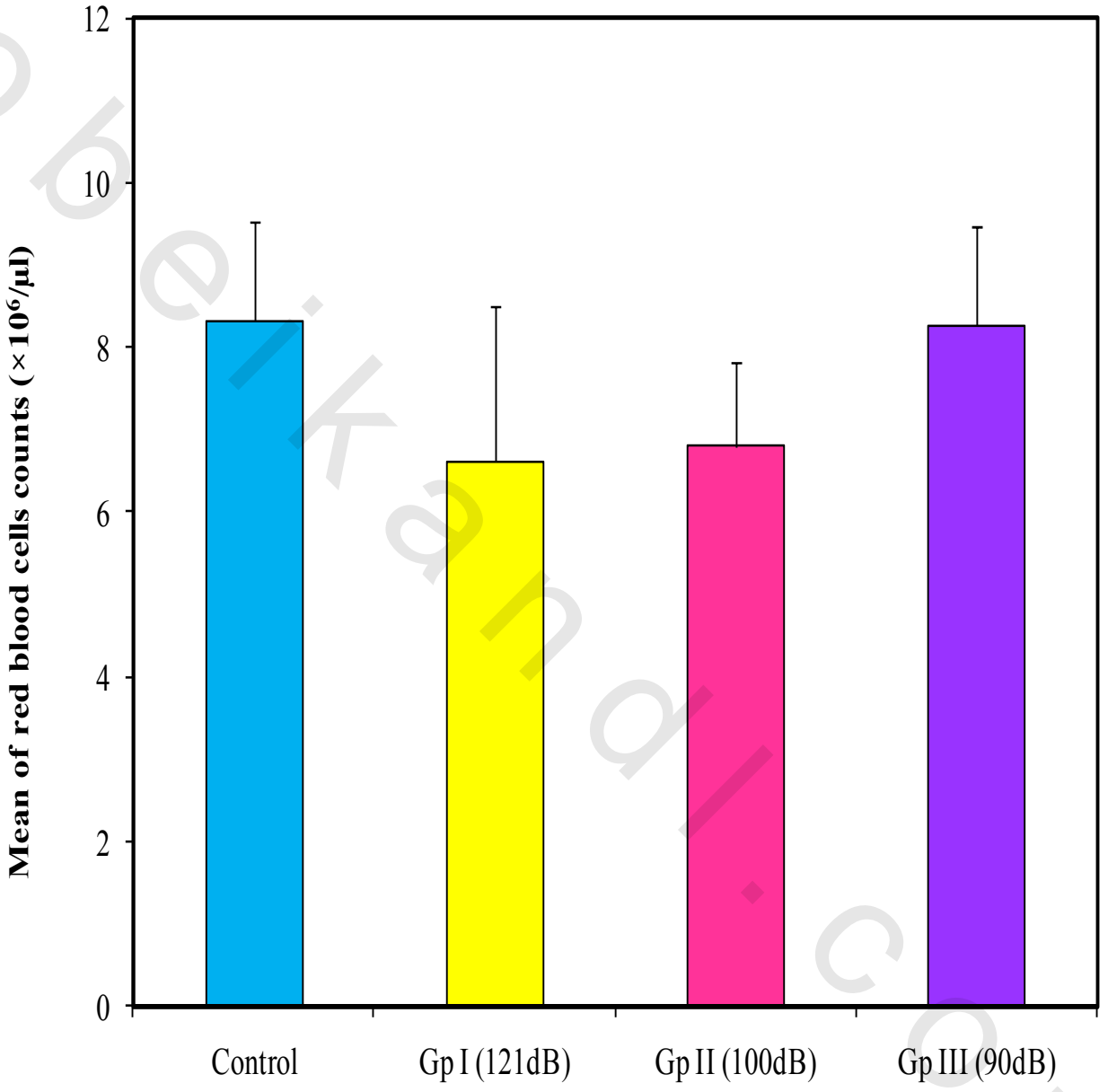


Figure (38): Effect of MRI noise exposure on RBCs count.

5.3.9. White blood cells counts:

WBCs counts were significantly increased for all groups exposed the acute noise as compared to control ($p < 0.008$). The statistical analysis of white blood cells counts are shown in Table (10) and Fig.(39).

Table (10): Statistical analysis of white blood cells counts (WBC) ($\times 10^3/\mu l$) of all different studied groups.

	Control (I) (n=10)	GB1 (II) (n=10)	GB2 (III) (n=10)	GB3 (IV) (n=10)
WBC ($\times 10^3/\mu l$)				
Min. - Max.	7.0 – 12.10	10.30 – 15.90	9.30 – 15.30	9.40 – 14.0
Mean ± SD.	9.50 ± 1.87	12.26 ± 1.70	11.19 ± 1.77	10.89 ± 1.33
% change from control		↑15.05	↑15.36	↑9.79
F	4.576*			
P	0.008*			
p_1		0.009*	0.188	0.346
p_2			0.573	0.359
p_3				0.984
Sig. bet. grps	I-II*			

F: F test (ANOVA)
p_1: p value for Post Hoc test (Scheffe) for comparing between control and each other group
p_2: p value for Post Hoc test (Scheffe) for comparing between GB1 with each of GB2 and GB3
p_3: p value for Post Hoc test (Scheffe) for comparing between GB2 and GB3
Sig. bet. grps was done using Post Hoc test (Scheffe)

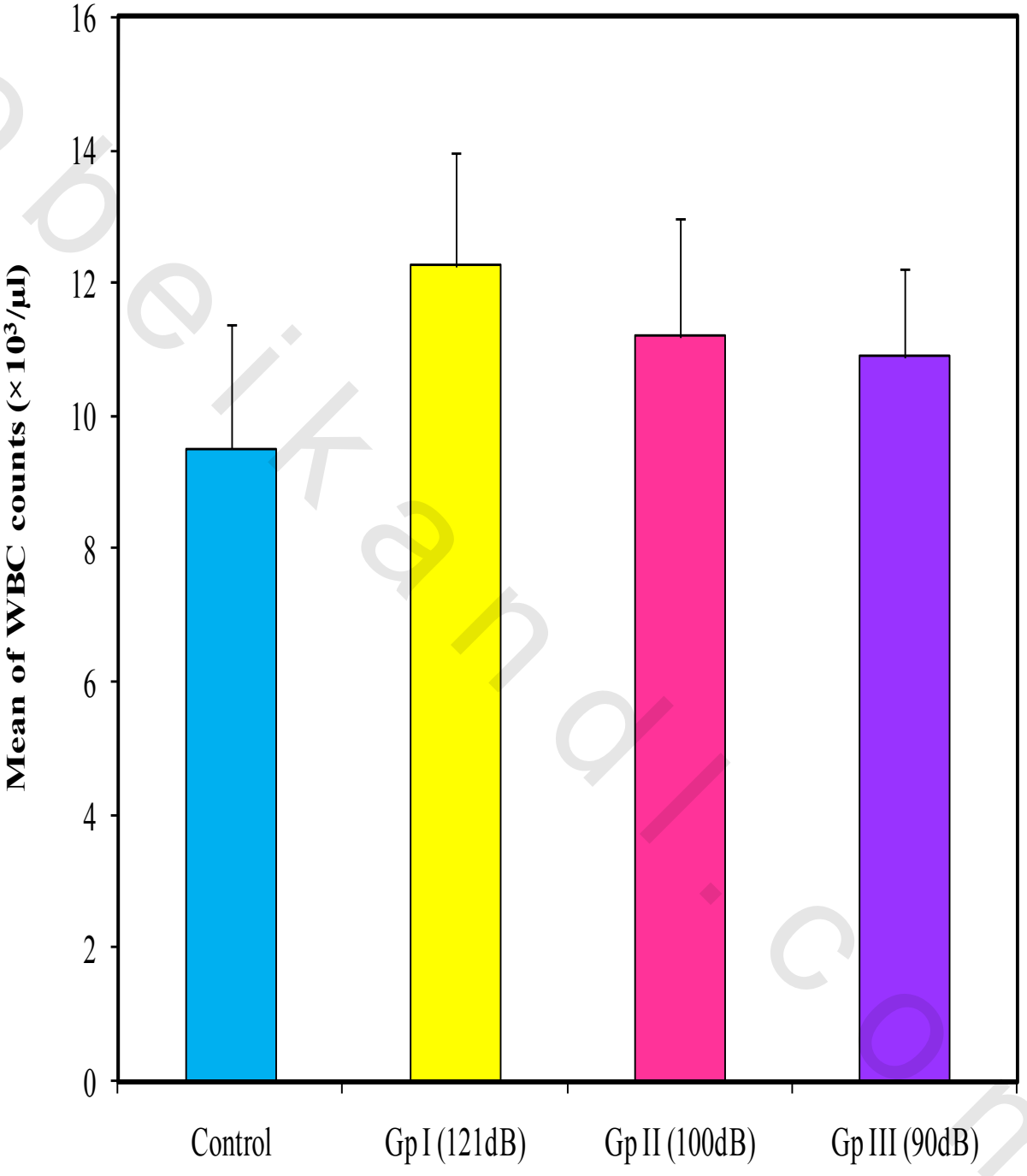


Figure (39): Effect of MRI noise exposure on WBCs count.

5.3.10. Platelets levels:

Platelets were significantly increased for groups exposed the acute noise as compared to control ($p < 0.001$). The statistical analysis of platelets are shown in Table (11) and Fig.(40).

Table (11): Statistical analysis of platelets($\times 10^3/\mu l$) of all different studied groups.

	Control (I) (n=10)	GB1 (II) (n=10)	GB2 (III) (n=10)	GB3 (IV) (n=10)
PLT ($\times 10^3/\mu l$)				
Min. - Max.	601.0 – 708.0	1063.0 – 1162.0	810.0 – 920.0	756.0 – 887.0
Mean ± SD.	653.70 ± 44.02	1125.50 ± 27.13	878.0 ± 39.89	873.27 ± 174.17
% change from control		↑72.17	↑34.31	↑0.72
F	268.452*			
P	<0.001*			
p_1		<0.001*	<0.001*	<0.001*
p_2			<0.001*	<0.001*
p_3				0.117
Sig. bet. grps	I-II*,I-III*,I-IV*,II-III*,II-IV*			

F: F test (ANOVA)
p_1: p value for Post Hoc test (Scheffe) for comparing between control and each other group
p_2: p value for Post Hoc test (Scheffe) for comparing between GB1 with each of GB2 and GB3
p_3: p value for Post Hoc test (Scheffe) for comparing between GB2 and GB3
Sig. bet. grps was done using Post Hoc test (Scheffe)

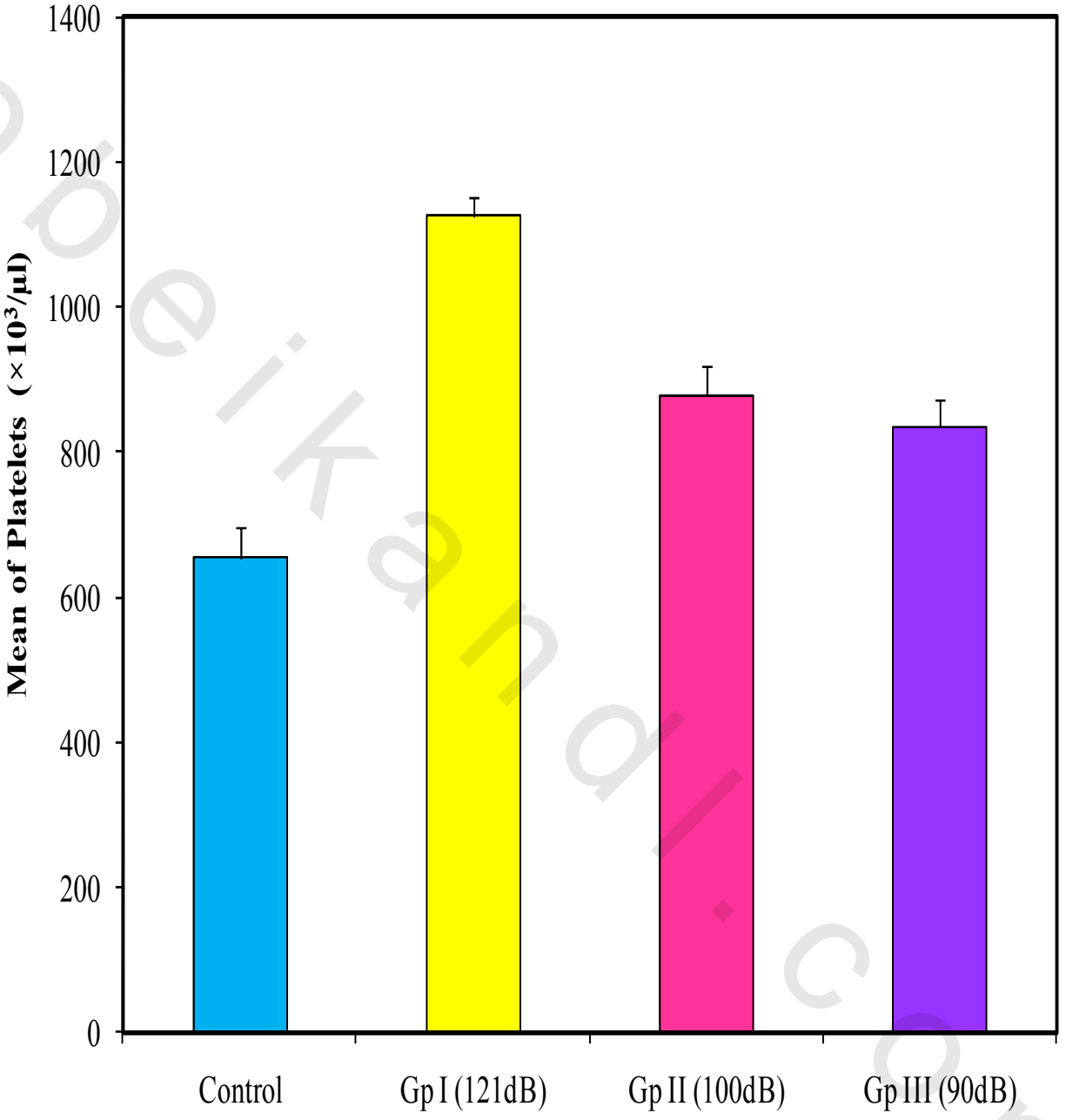


Figure (40): Effect of MRI noise exposure on platelets levels.

5.3.11. Effect of noise on malondialdehyde (MDA) in brain of mice:

The individual data and the mean values ± S.D for malondialdehyde in all different studied groups are shown in Table (12).The statistical analysis of these results represented in Table (13) and Fig.(41).

It was demonstrated that the activity of MDA in brain ranged from 0.04-0.18 with mean value of 0.08 ±0.05 nmol/ mg protein in control group. On the other hand, in mice exposed to 121 dB (GB1) MDA level ranged from 0.11 – 0.36 with mean value of 0.21 ± 0.08 nmol/ mg protein which showed significant increase as comparison to control group ($P < 0.001$). In mice exposed to 100 dB (GB2) the MDA level ranged from 0.03 – 0.15 with mean value of 0.09 ± 0.03 nmol/ mg protein, there is no difference in this group as compared to control group ($P < 0.001$). In group exposed to 90 dB (GB3), MDA level was in the range of 0.05 – 0.33 with mean value of 0.15 ± 0.10 nmol/ mg protein which is significantly higher than control group ($P < 0.001$).

Table (12): Malondialdhyde levels (as n mol/ mg protein) of mice brain tissues to studied of experimental groups. (GB1 exposed to 121 dB, GB2 exposed to 100 dB, and GB2 exposed to90 dB) in comparison with control group.

No.	Control	GB1	GB2	GB3
1	0.110	0.222	0.127	0.096
2	0.178	0.365	0.083	0.102
3	0.055	0.192	0.062	0.056
4	0.073	0.114	0.076	0.077
5	0.035	0.349	0.099	0.325
6	0.081	0.166	0.030	0.066
7	0.141	0.211	0.059	0.193
8	0.049	0.153	0.068	0.052
9	0.057	0.166	0.146	0.250
10	0.067	0.196	0.096	0.246
Min.	0.04	0.11	0.03	0.05
Max.	0.18	0.36	0.15	0.33
Mean	0.08	0.21	0.09	0.15
SD.	0.05	0.08	0.03	0.10

Table (13): Statistical analysis of malondialdehyde (MDA) levels (as n mol/ mg protein) of mice brain tissues of all different studied groups.

	Control (I) (n=10)	GB1 (II) (n=10)	GB2 (III) (n=10)	GB3 (IV) (n=10)
MDA levels (as n mol/ mg protein)				
Min. - Max.	0.04 – 0.18	0.11 – 0.36	0.03 – 0.15	0.05 – 0.33
Mean ± SD.	0.08 ± 0.05	0.21 ± 0.08	0.09 ± 0.03	0.15 ± 0.10
F	7.713*			
P	<0.001*			
p_1		0.003*	1.000	0.284
p_2			0.003*	0.229
p_3				0.284
Sig. bet. grps	I-II*,II-III*			

F: F test (ANOVA)
p_1: p value for Post Hoc test (Scheffe) for comparing between control and each other group
p_2: p value for Post Hoc test (Scheffe) for comparing between GB1 with each of GB2 and GB3
p_3: p value for Post Hoc test (Scheffe) for comparing between GB2 and GB3
Sig. bet. grps was done using Post Hoc test (Scheffe)
*: Statistically significant at $p \leq 0.05$

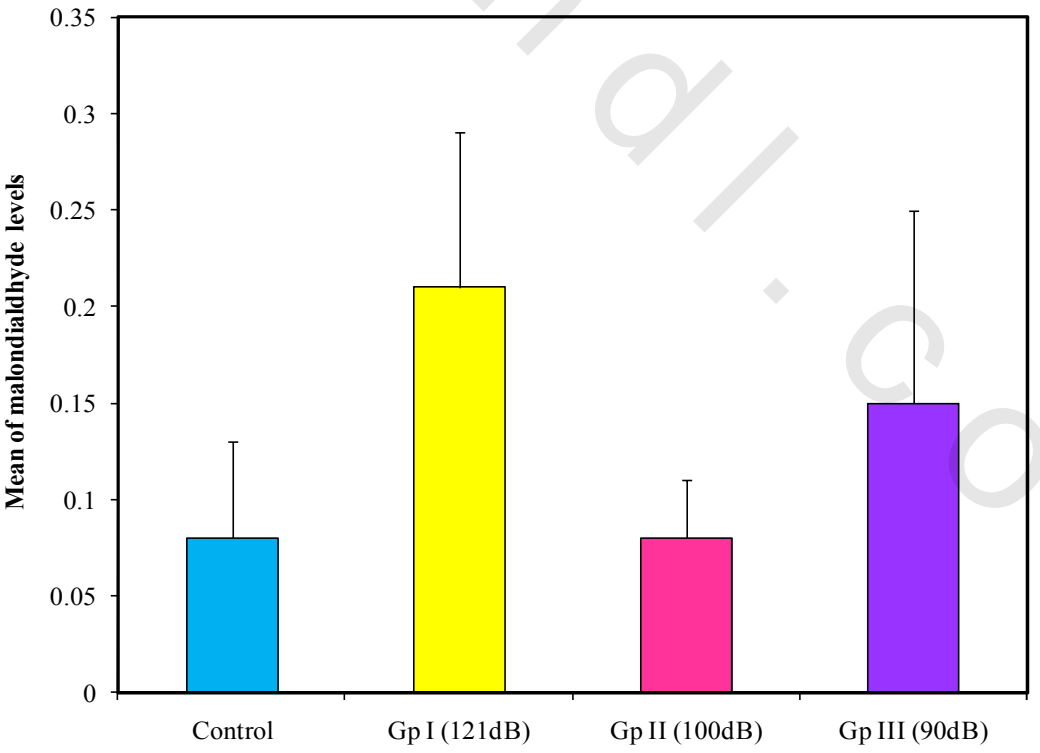


Figure (41): Effect of MRI noise exposure on MDA levels.

5.3.12. Effect of noise on superoxide dismutase (SOD) activity in mice brain:

The individual data and the mean values ± S.D for superoxide dismutase (SOD) activity in all different studied groups are shown in Table (14).The statistical analysis of these results represented in Table (15) and Fig.(42).

It was demonstrated that SOD activity in all exposed to MRI noise in the brain tissues were significantly decreased as compared to control group ($p < 0.001$). SOD level in control group was in the range of 0.002 – 0.006 with mean value of 0.0045 ± 0.001 IU / mg protein. SOD level in mice exposed to 121 dB (GB1) SOD level ranged from 0.002 – 0.003 with mean value of 0.0017± 0.00067 IU / mg protein which is significantly decreased as compared to control group ($P < 0.001$). In mice exposed to 100 dB (GB2) the SOD level ranged from 0.001 – 0.004 with mean value of 0.0018±0.00092 IU / mg protein. In group exposed to 90 dB (GB3), SOD level was in the range of 0.001 – 0.003with mean value of 0.0021± 0.00073 IU / mg protein. It is clear that, there was significantly reduction in SOD level in all experimental groups as compared to control group ($P < 0.001$).

Table (14): Superoxide dismutase (SOD) activity (as IU / mg protein) of mice brain tissues of experimental groups. (GB1 exposed to 121 dB, GB2 exposed to 100 dB, and GB2 exposed to90 dB) as comparison with control group.

No.	Control	GB1	GB2	GB3
1	0.005	0.003	0.004	0.003
2	0.004	0.001	0.002	0.002
3	0.004	0.001	0.001	0.002
4	0.006	0.002	0.001	0.001
5	0.005	0.002	0.001	0.002
6	0.006	0.001	0.001	0.002
7	0.005	0.002	0.002	0.001
8	0.006	0.002	0.002	0.003
9	0.005	0.001	0.002	0.002
10	0.006	0.002	0.002	0.003
Min.	0.002	0.001	0.001	0.001
Max.	0.006	0.003	0.004	0.003
Mean	0.005	0.0017	0.0018	0.0021
SD.	0.001	0.00067	0.00092	0.00073

Table (15): Statistical analysis of Superoxide dismutase (SOD) activity (as IU/mg protein) of mice brain tissues of all studied groups.

	Control (I) (n=10)	GB1 (II) (n=10)	GB2 (III) (n=10)	GB3 (IV) (n=10)
Superoxide dismutase (SOD) levels (as IU / mg protein				
Min. - Max.	0.004 – 0.006	0.001 – 0.003	0.001 – 0.004	0.001 – 0.003
Mean ± SD.	0.005 ± 0.001	0.0017 ± 0.00067	0.0018 ± 0.00092	0.0021 ± 0.00073
F	45.514*			
P	<0.001*			
p_1		<0.001*	<0.001*	<0.001*
p_2			0.994	0.731
p_3				0.866
Sig. bet. grps	I-II*, I-III*, I-IV*			

F: F test (ANOVA)
p_1: p value for Post Hoc test (Scheffe) for comparing between control and each other group
p_2: p value for Post Hoc test (Scheffe) for comparing between GB1 with each of GB2 and GB3
p_3: p value for Post Hoc test (Scheffe) for comparing between GB2 and GB3
Sig. bet. grps was done using Post Hoc test (Scheffe)

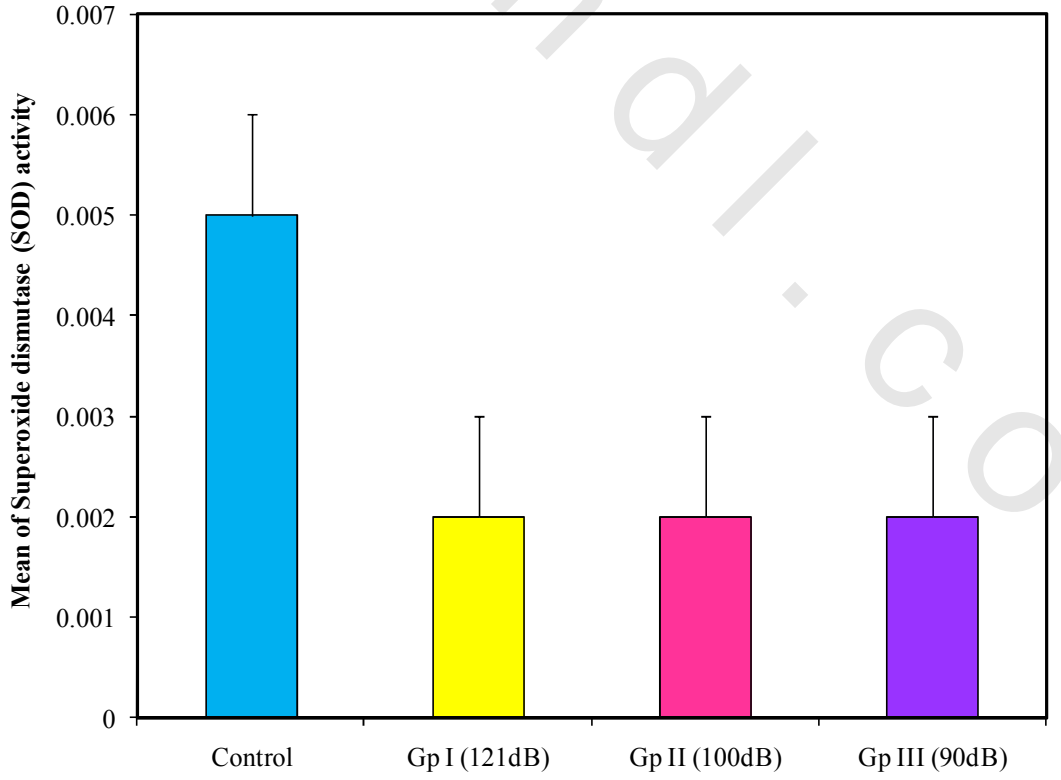


Figure (42): Effect of MRI noise exposure on Superoxide dismutase (SOD) activity.