

BIO-EFFECTS OF AIRCRAFT NOISE AND ANTIOXIDANT ACTIVITY OF OMEGA3 ON MICE BRAIN

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ABSTRACT

In order to investigate the non-auditory bio-effects of aircraft noise exposure on brain, mice in soundproof chambers were exposed to a previously recorded aircraft-related noise signal for a duration of 1,2 or 3 weeks. For comparison, unexposed control mice have been used. The work plan includes aircraft takeoff and landing noise which adjusted to a level of 100 dB for the 4 experimental groups for 2 hours daily. The recorded noise was analyzed using Audacity software program. The levels of blood glucose and cortisol in serum were measured. Morphological changes of brain tissue were observed by light microscope. The results showed that, Takeoff frequency ranged from 3-4 KHz at 100dB, while dominant frequency for landing was 2-3 KHz. Noise of 100 dB was found to cause significant increases in plasma glucose and cortisol for exposed mice as compared to no noise exposure. Administration of omega 3 for three weeks causes a significant improvement in plasma glucose and cortisol. Histopathological changes in brain of mice exposed to aircraft noise revealed some vesicles and mild to moderate apoptosis and pyknosis of the nuclei. In conclusion the present study determined that high intensity noise has a direct and clear effect on physical, biochemical values and thus noise exposure should be well organized.

KEY WORDS:

Aircraft noise, Stress, Omega 3, Bloodglucose , Cortisol, Histopathological changes.

1. INTRODUCTION

Noise is an unavoidable part of our daily lives and has increasingly become a major burden on the quality of lives. Noise, has been shown to produce a number of physiological, biochemical, and neurochemical rejoinder in both human and animals. High levels of noise can be associated with impairment of ability to concentrate [1, 2]. Aircraft noise has been shown to influence cardiovascular health in adults, and there is some evidence that the average blood pressure was significantly higher in the group with higher noise exposure [3]. Both long-term as well as heavy studies of animals have provided biological mechanism and plausibility for the theory that exposure to environmental noise affect the central nervous system and causes manifest diseases. The brain, the key organ that interprets and be interrogated to potential stressors, recognizes the sound levels and differentiate the stress levels. It reacts within split seconds timing to instruct the rest of the body in how to adjust to this stressful situation by the release of cascade of hormones [4,5].Investigation the physiological effects of aircraft noise under the condition of laboratory, indicated that medium and short term noise exposure (<15day) could produce anxiety on rats, however, long term noise exposure (>30d) could produce depression or com or bid anxiety and

depression on rats. However, the results of neurotransmitters showed that, Norepinephrine, NE and 5-hydroxytryptamine, 5-HT concentration of experimental group increased at the beginning, then decreased with the time [6]. Furthermore, noise exposure for 15 days was reported to affect serotonin and dopamine levels in the hippocampus and produced recognition memory failure in rats[2].

The alteration in the levels of plasma corticosterone after different durations of noise exposure have been studied by Samson et al [5] to analyze their role in combating time –dependent stress effects of noise. Broadband white noise of (100 dB) exposure to male albino rats significantly increased the levels of plasmacorticoster one after different duration (acute, 1 d; sub-acute, 15 d; chronic, 30 d) of noise exposure. Also, association between noise exposure and plasmacorticoster one levels was studied by Vitaleet al [7]. A 10 minutes exposure to noise stress resulted in increased corticosterone levels in mice plasma.

Observing the morphological changes in neuronal cells can assess the effects of noise. The detrimental effects of noise stress on both deterioration of epithelial cells and apoptosis of stromal cells in the brain have been well documented in several studies[7,8]. The findings of these studies indicated that aircraft noise exposure leads to the neurons of temporal lobe began to apoptosis, synapse morphology had changed, synaptic cleft of hippocampus and the temporal lobe area was vague, chromatin aggregated and a synaptic vesicle was not clear [7]. Di et al[9] exposed rats to previously recorded aircraft-related noise at 75 and 80 dB for 65d. The results showed that the neuron and synaptic structures of the temporal lobe have signs of damage after aircraft noise of 80 dB exposure for 65 days.

Due to the high demand for energy from the brain, it would be susceptible to oxidation. Also neurons are sensitive to oxidative stress, as it is considered responsible for the occurrence of many neurological diseases and cell death [10]. Omega-3 polyunsaturated fatty acids block apoptotic neural cell death. Therefore, they are considered a strong protective against deterioration of acute and chronic neurodegenerative. They reduce pro-inflammatory mediators, increase the production of anti-inflammatory mediators and reduce susceptibility to lipid peroxidation and oxidative stress in neural cells[11].

None of the aforementioned studies studied physical analysis of aircraft noise. In this study, actual aircraft noise was played back to laboratory mice. Analysis the frequency of the noise, blood glucose level, cortisol level in serum and cell morphology of the brain tissue with and without omega -3 were studied.

2. Materials and Methods

2.1 Materials

The work plan included, recording takeoffs and landings of aircraft from the following link www.soundjay.com as a source of noise and then it was run through loudspeaker inside anechoic chamber.

Anechoic chamber:

Figure 1 illustrated schematic diagram of anechoic chamber in which the mice were placed to be exposed to the noise. Homemade 70x70x70cm³ plywood box as anechoic chamber was made, lined with polyurethane foam from all sides. The chamber contains illumination and ventilation system. 15 W RMS loud speaker was installed on one side of the anechoic chamber, driven by the power amplifier (Model FUS- 432 RFL (Made in China) that can transmit the recorded specified

noise with the required dB value (Fig 1). Noise level was measured by a sound level meter (Lutron electronic enterprise Co, LTD.) Model SL- 4010, 35to 130 dB, 3 ranges, Data hold and an instrument which responds to sound in approximately the same way as the human ear, where it gives repeatable measurement of sound level. The recorded noise was analyzed using Audacity software program (version 2.0.5) <http://audacity.sourceforge.net>. Repeating area of the sound waves with the full characterization was obtained through the software analysis.

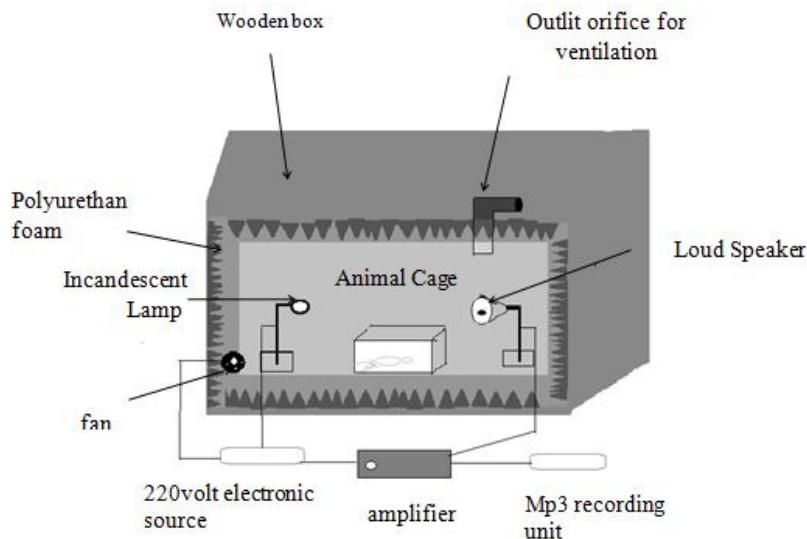


Figure1. Schematic diagram shows anechoic chamber

2.2 Animals

Use of experimental animals in this study was carried out in accordance with the ethical guidelines of the Medical Research Institute, Alexandria University, (Appendix2 Guiding principles for Biomedical Research Involving Animals, 2011).

Animal Study Groups:

Eighty mice weighted 25-30 gm was purchased from the Experimental Animal Center of Medical Research Institute. The mice were kept in comfortable cages, well air conditioned, light system of 12 hr day/ 12 hr dark, food and water were allowed freely. Mice were divided into three main groups. The division process was completely at random.

1-Control group was divided into two subgroups (10 mice each) as follow:

- Control group (A1) was remaining in the anechoic chamber for two hours daily without exposure to any sound.
- Control group (A2) was administered oral supplementation of daily dose of omega-3 fatty acid 100 mg/kg body weight, then remaining in the anechoic chamber for two hours daily without exposure to any sound.

2-Experimental group (B) consists of 30 mice was divided into three subgroups (10 mice each).

These three subgroups exposed to aircraft noise (takeoff and landing) at intensity of 100dB for 2 hours daily as follows:

- GB1 for one week.
- GB2 for two weeks.
- GB3 for three weeks.

3-Experimental group (C) consists of 30 mice was divided into three subgroups (10 mice each).

These three subgroups administered oral supplementation of daily dose of omega-3 fatty acid 100 mg/kg body weight, then exposed to aircraft intensity of 100dB as follows:

- GC1 for one week.
- GC2 for two weeks.
- GC3 for three weeks.

2.3 Biochemical Measurements:

2.3.1 – Glucose level measurement

The glucose assay uses the glucose oxidase- peroxide reaction for determination of glucose concentrations by generation of a pink dye with an optimal absorption at 514nm [12]. Blood samples were taken from control and experimental groups during dissection. For plasma glucose analysis, the blood samples were collected in the gray tubes which contain sodium fluoride and potassium oxalate then kept in refrigerator at -20 °C until analysis.

2.3.2- Cortisol level measurement

Due to the circadian rhythm of cortisol levels in serum, the sample collected in the morning. The trunk blood was centrifuged at 400 rpm for 30 minutes at 4 oC, serum collected using standard sampling tubes, and stored at -8°C until assayed. Before the determination of corticosterone (CORT), serum was diluted 3:1 in assay buffer and assayed in duplicate using commercially available Elecsys2010 (Sarstedt, Numbrecht, Germany).

2.4- Histopathological studies:

A cryosectioning of brain at low temperature -60oC, (10µm) thick using SLEE MAINZ; type MEV) cryostat with a stainless steel knife was used. The section was fixed at 10% formaldehyde then washed in dis.H2o. The nuclear fast red was used as a counter stain, then the slide left to dry and mounted by glycerine-gel and covered by cover slip to be observed under light microscope (BX41; Olympus America Inc). A photograph with digital compact camera (CAMEDIA C -7070, U- CMAD3, Olympus Company with high performance 7.1 megapixel colored camera was taken.

3-Results

3.1-Analysis of aircraft noise:

3.1.1 -Band selection and expansion of takeoff and landing noise

Figure 2 (a,b,c&d) shows amplitude- time plots in both , time – unexpanded and time- expanded modes respectively. Amplitude is measured in arbitrary units, and the time in sec. The unexpanded plot contains screenshot of the entire recorded takeoff signal sound, with a vertical red line showing where the time-expanded sections were selected. The expanded waveform shows random variation in amplitude.

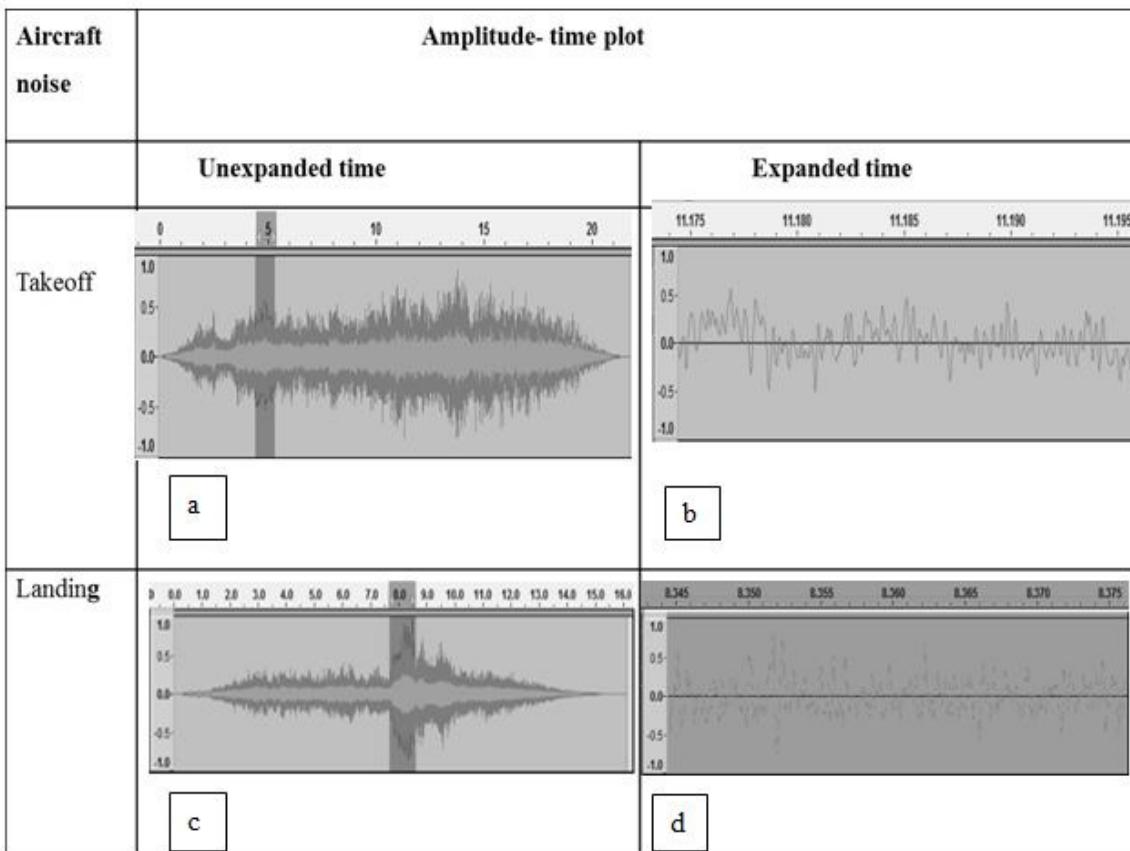


Figure.2.(a,b, c &d): Take off noise amplitude-time plot in unexpanded and expanded mode

3.1.2. Spectrum of takeoff and landing:

A sound spectrum displays the different frequencies present in a sound. It represents a short sample of a sound –in terms of the amount of vibration at each individual frequency. Figure 3 (a&b) shows spectrum analysis of takeoff and landing noise. The X- axis represents the sound frequency in Hz, while the Y-axis represents the sound intensity in dB. As shown from the figure the highest frequency ranged from 3to 4 KHz and 2to 3 KHz for takeoff and landing respectively.

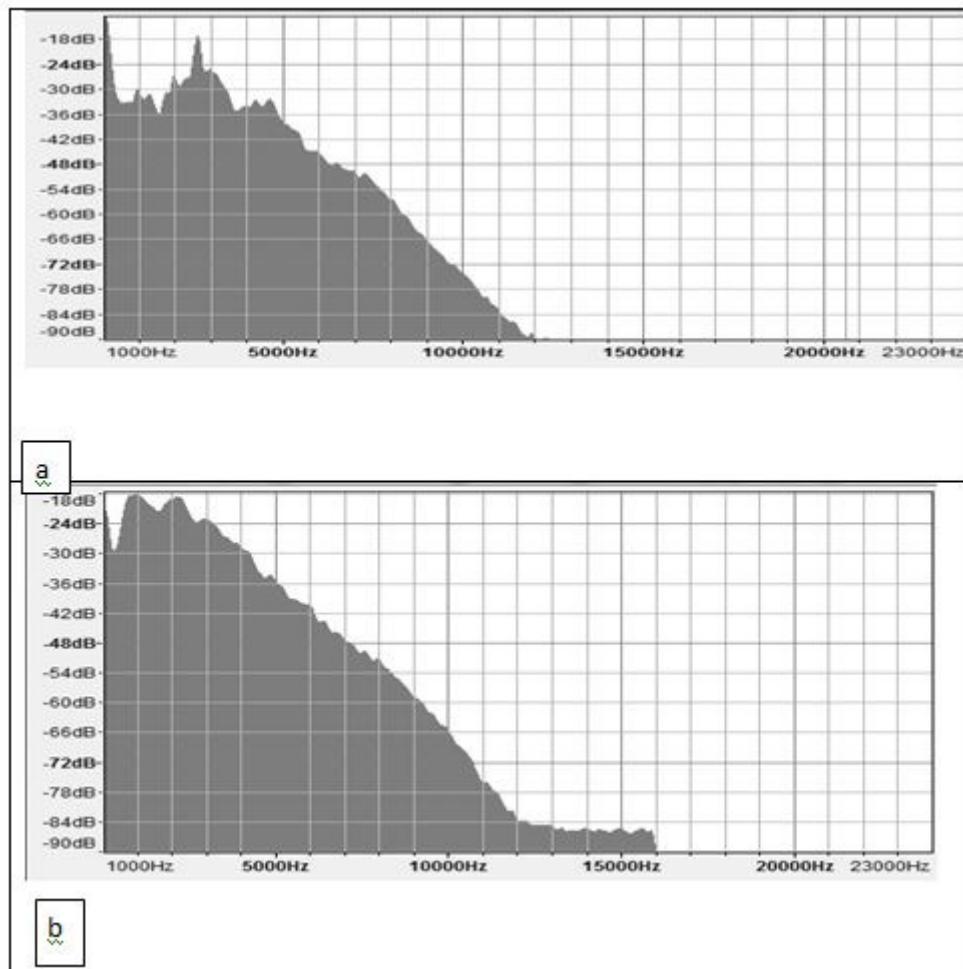


Figure 3 (a&b):Spectrum analysis for takeoff and landing of aircraft noise

3.1.3-Cepstrum analysis and standard Auto correlation:

Cepstrum is an analysis function with many application in signal processing. It is used as a complementary tool to spectral analysis, it helps identifying items which not readily identified by spectral analysis. Takeoff and landing cepstrum analysis are shown in figure (4a&c) for takeoff and landing noise . The cepstrum diagram shows sporadic very high spikes of sound that are closed to aircraft and or workers speech and any movement are separated to the left of the diagram as a characteristic band, while the constant annoying sound appears at the rest of the axis. As it is already known that autocorrelation measures, to what extent the sound repeats itself. In other words how many identical frequencies in the used sound sample. From the autocorrelation figure (4b&d) of the selected takeoff and landing noise band, the higher level sound intensity is separated at the left side of the plot (unrepeated), while the continuous sound repeated in the rest of distinct bands.

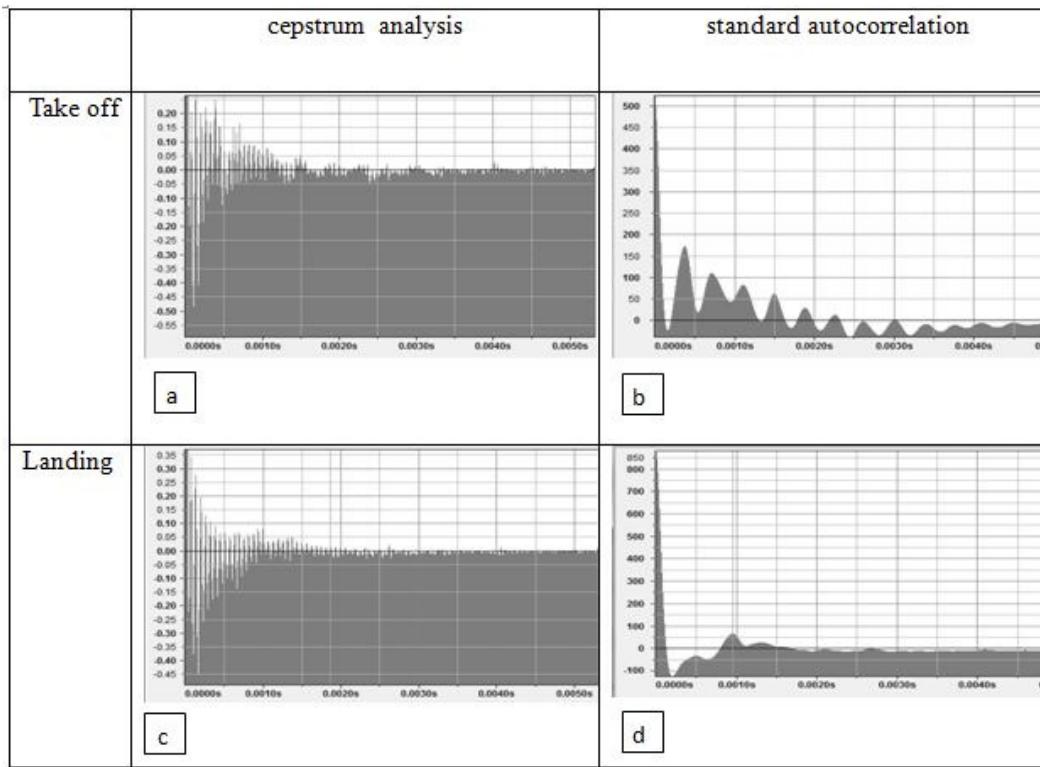


Figure 4.(a,b,c & d) : Takeoff and landing cepstrum analysis and standard autocorrelation

3.2- Biochemical studies:

3.2.1- Blood glucose level

Blood glucose level (mg/dl) in all groups exposed to takeoff and landing noise with or without omega 3 were represented in Figure 5. It is clear that blood glucose, were significantly increased in its concentration for groups exposed to noise as compared to control ($p < 0.05$). Groups that were administered omega-3 before exposure to the sound did not give any significant reduction in the blood glucose level with the exception of group administered omega 3 for three weeks then exposed to takeoff and landing sound. Blood glucose of this group significantly decreased from (212.4 ± 22.8 mg/dl to 122.4 ± 15.9 mg/dl) ($p < 0.05$) as compared to group exposed to takeoff and landing noise without omega.

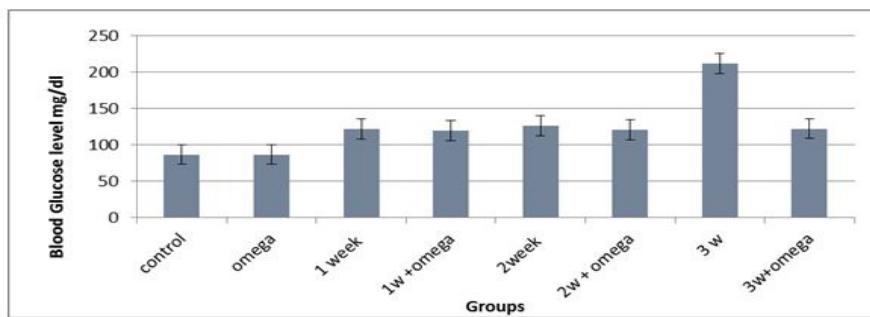


Figure 5: Effect of exposure to aircraft noise on blood glucose level

3.2.2- Cortisol level

Figure 6 shows the relationship between the exposure of control and experimental groups to aircraft noise at 100dB intensity with different durations and the average concentration of plasma cortisol level. Data explained that there was significant difference in cortisol level with different groups when compared to control group ($p < 0.05$). Groups administered omega 3 show no significant decrease in cortisol level even after 3 weeks. Figure 7 shows linear relation between cortisol and blood glucose level, from this relation increase in blood glucose could be calculated from equation $Y = 38.7x - 72.7$ where y is blood glucose and x is cortisol level with a correlation coefficient of $r=0.8098$.

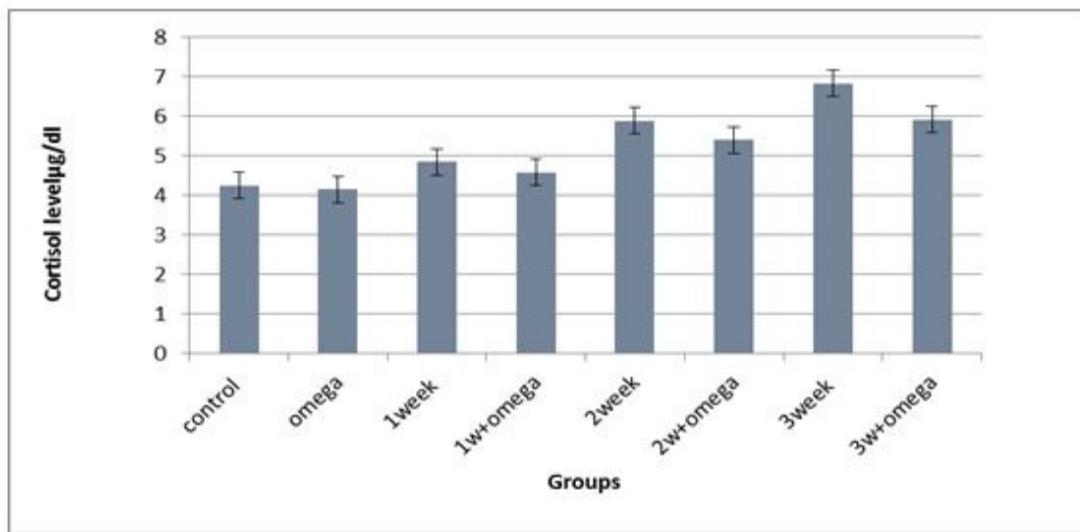


Figure. 6: Relationship between cortisol level and noise exposure duration in different group

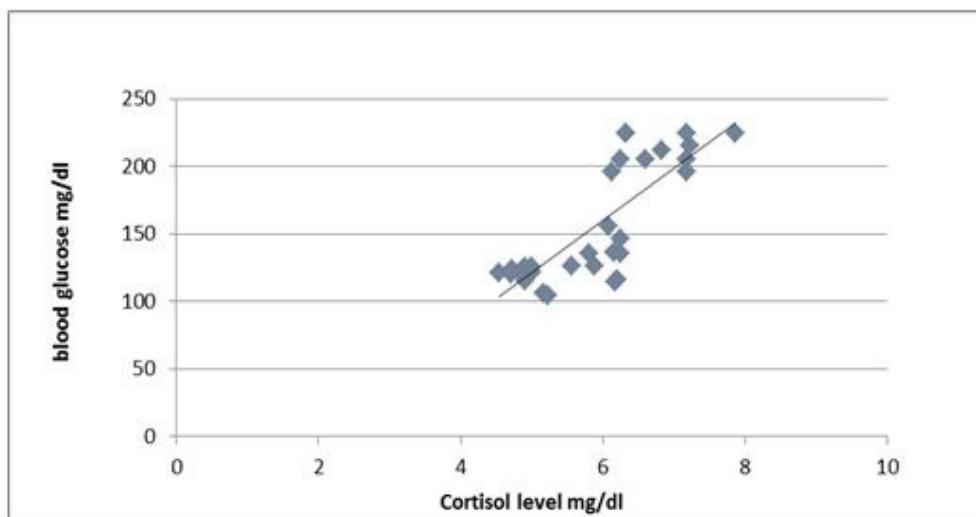


Figure 7: Linear relation between cortisol and blood glucose

3.2.3-Histopathological study

The histopathological changes in brain of mice exposed to different duration one, two and three weeks with or without omega 3 to aircraft noise in comparison with control group were shown in figures (8-11). Light micrograph showed that mild to moderate vesicles, neurons of experimental group exposed to noise for three weeks began to pyknosis and apoptosis.

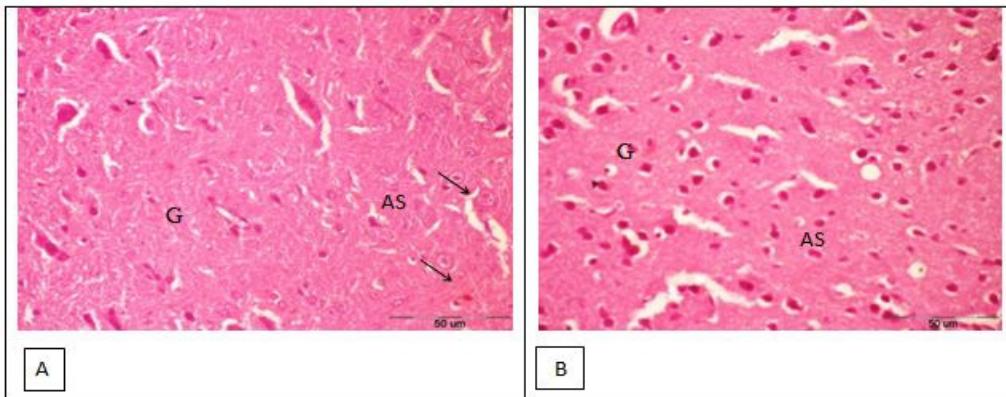


Figure (8) photomicrograph of control group reveals normal brain tissue formed of glial tissue (G) and astrocytes (arrows AS). A: Normal brain, B:Normal brain with omega 3.

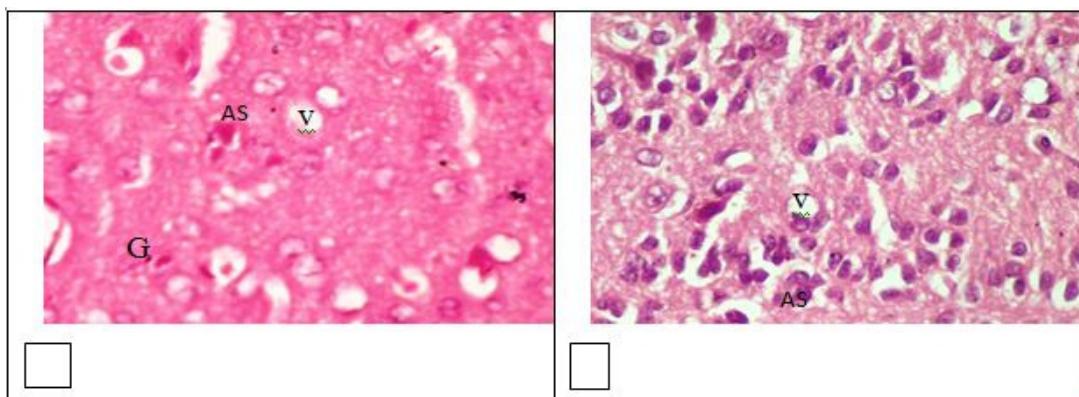


Figure 9: Photomicrograph of group exposed to noise for one week.A: without omega, B: with omega) reveals normal glial tissue (G) and astrocytes (AS) with some vesicles (V).

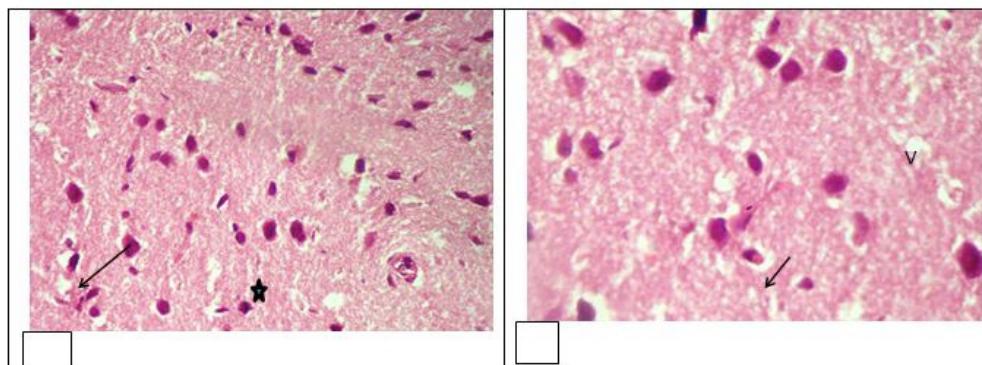


Figure 10: Photomicrograph of group exposed to noise for two weeks. A: without omega, B: with omega) reveals apoptotic nuclei (arrow) and pyknoticcell (*) with many vesicles (v).

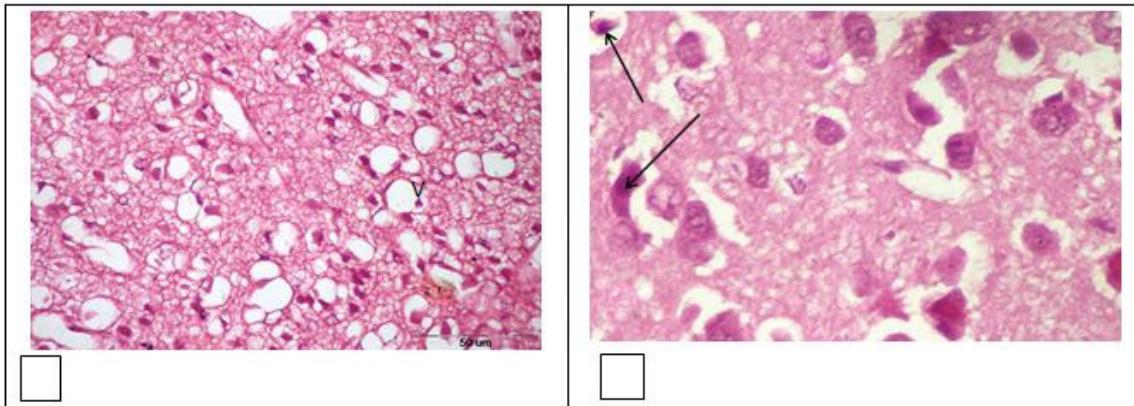


Figure 11: Photomicrograph of group exposed to noise for three weeks. A: without omega, B: with omega) showing fragmentation of nuclear membrane with pyknosis of nuclei (arrow) and with numerous vesicles (V).

4. DISCUSSION

Occupational noise is one of the main annoying stressor to which a large population is being exposed for a long duration daily. Many studies have reported that exposure to aircraft noise causes many health problems for populations working in or near airport such as cardiovascular diseases, lack of concentration, fatigue, heart trouble, plasma viscosity, high glucose level and reduced motor efficiency [13].

Noise usually consists of sound signals with a broad spectrum, including both low frequency and high frequency region [14]. A disreputable example of low frequency noise is the noise generated by the take-off roll of aircraft on the runway, this type of aircraft noise known as aircraft groundnoise[15].

The present study was undertaken to analyze noise signals and spectrum of takeoff and landing of aircraft. According to figure 2 (a,b,c, and d) shape of aircraft noise has a repeated signals with different intensities and different frequencies. The spectrum of noise emitted from takeoff has the highest levels in the frequency band ranged from 3 KHz - 4 KHz at 100dB and gradually decrease above this frequency. The spectrum of noise emitted from landing has the highest levels in the frequency band ranged from 2-3 KHz at 100dB and fall off more sharply above this frequency at 4KHz to 83.58dB. This analysis is a good agreement with Berglund et al [16] study which revealed that, although the spectra are often not linear, aircraft groundnoise spectra measured in communities near an airport are nearly linear at a frequency ranged from 1-2 kHz and then fall off more steeply above this frequency. Starting with the effect of noise on blood glucose level, noise exposure is a stressful condition and stress is known to signal the body to raise blood glucose level in order to generate energy to respond to the stress [17]. The relationship between the effect of noise and its impact on blood glucose has been studied. The results showed that, an elevation of the plasma blood glucose for all groups' noise exposure as compared to control. The study also showed that the three weeks noise exposure group had a highest level of plasma blood glucose than that of control, one week and two weeks with or without omega 3 supplement as shown in figure (5). Blood glucose level also increased in the study held by Emily and Mark[18] their results revealed that, there was an increase in blood glucose level after exposure to aircraft noise of a maximum level 105 dB for 3 seconds. Building upon Muayad et al [19]

studies, the effects of acute and chronic loud noise exposure ranged from 80-100 dB was found to cause high levels in plasma blood glucose as compared to no noise exposure.

Cortisol plays a vital role in health maintenance. But in order to do so, circulating levels must be maintained within a narrow range. If levels less than optimal, signs and symptoms of adrenal weariness occur; if levels increased and remain above optimal for a period of time, signs and symptoms of metabolic syndrome occur [20].

Rising in cortisol levels have been found in humans and animals that exposed to aircraft noise or road traffic noise [21]. The present study observed the effect of noise on cortisol as stress hormone, where there was an elevation of cortisol for all groups' noise exposure as compared to control. The study also showed that the three weeks noise exposure group had a highly level of cortisol in comparison to control, one week and two weeks with or without omega 3 supplement as shown in figure (6). The aforementioned results are similar to those reported by Hartono[22] experiment where group exposed to aircraft noise at an intensity level of 92.29 dB the noise acted as a stressor, resulting in a higher increase in mean cortisol level than was the case with aircraft noise at an intensity level of 71.49 dB. Furthermore, Cheng and Ariizumi[23] who did an experiment with BALB/c mice at noise intensity level of 90 dB, with a length of exposure of five hours per day for four weeks. Mean of cortisol level of treated group exposed to noise was 4.25 µg/dl, which was higher than that of the control group of 1.60 µg/dl.

There is a strong relationship between cortisol and blood glucose levels as shown in figure (7). Under stressful condition, sufficient concentrations of cortisol mobilize glucose into the bloodstream via gluconeogenesis, reduced insulin sensitivity, amino acid motivation and protein catabolism. Cortisol obtains quick glucose for the body to use in times of stress. Also, in stress time cortisol gets rapid glucose for the body to use while reduce insulin effects, so blood glucose levels becomes high and insulin unable to perform its regular function of maintaining normal glucose levels[24]. On the other hand, according to this relationship, diet plays a main role in adrenal health. Poor eating habits and food choices not only leave the body without essential nutrients but also impede or create a burden on the stress response, digestive, energy production and other homeostatic systems. There are specific dietary guidelines that form the other side of the therapeutic effect [25]. In the present study omega 3 was used as diet supplements, the results indicate that blood glucose and cortisol decreased after intake of omega for three weeks as shown in figure (5,6). This study is a good agreement with Delarue, et al [26] study, which evaluated the effect of n-3 fatty acids on the hypothalamic-pituitary-adrenal (HPA) axis. Their results indicated supplementation with 7.2 g of fish oil per day, the cortisol response to stress was significantly reduced after 3 weeks.

5. CONCLUSION

Depending on the present study, aircraft noise may lead to the negative effect on blood glucose level and cortisol level, particularly in long term noise exposure. Also, there was a linear relationship between circulating cortisol levels with blood glucose level due to depletion of insulin resistivity. Moreover, the study indicated that aircraft noise exposure leads to histological changes in neuronal structures of the mice brain. Increased natural antioxidant and neuro protective like Ginkgo biloba or Panax ginseng intake to reduce the oxidant stress produced by noise. It must be taken in consideration, increased improvements and control methods from noise based on intensity and frequency levels.

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