Al-Nahrain Journal of Science
ANJS, Vol. 27(2), June, 2024, pp. 127-132

The Effect of Acute and Chronic Noise Stress Exposure on some Physiological and Immunological Parameters in Male and Female Adult Mice

Zainab A. A. Al-Shammary[1,*], Sally A.K.[1], Zainab Sameer Sabti[2], Jamela Jouda[3], Sabah A. Hameid[4]

[1]Medical Physics Department, College of Science, Alnahrain University, Jadiriya, Baghdad, Iraq.
[2]Criminal Evidence Department, College of Science, Al-Nahrain University, Jadiriya, Baghdad, Iraq.
[3]Department of Biology, College of Science, Mustansiriyah University, Baghdad, Iraq.
[4]Biology Department, College of Science, Alfarabi University, Baghdad, Iraq.

Article's Information

Received: 15.01.2024
Accepted: 14.05.2024
Published: 15.06.2024

Keywords:
Chronic Noise
Stress
IgG, and IgM

Abstract

Noise, the most commonly encountered stressor in our daily lives, affects the body in different ways depending on its frequency, intensity, and duration. Thus, this study aimed to demonstrate the effect of noise stress exposure in different durations on the immunological status of female and male adult mice. Sixty female and male adult mice (Balb / C) were used in this. Fifty mice were exposed to noise stress at 90 degrees for four hours per day, and 10 mice (5 males and 5 females) were left without exposure to stress which was used as a control group. Exposure stress mice were divided into five groups (n = 10, 5 males and 5 females); Group1 was exposed to noise stress for only 1 day, Group2 for 8 days, Group3 for 14 days, Group4 for 21 days, and Group5 for 30 days. The blood sample was collected and used for immunoglobulins, glucocorticoids and WBCs indices tests. Results showed that there was a highly significant increase in glucocorticoids for all periods after noise exposure. WBC count, the level of IgG and IgM were highly significant in mice that were exposed to noise stress during 1, 8, 14, and 21 days, while there was no significant difference in the 28-days stress group compared to control. In conclusion, the levels of immunoglobulins levels after 28 day of noise stress exposure could completely differ from the short-term exposure.

http://doi.org/10.22401/ANJS.27.2.13
*Corresponding Author: zainab.ali@nahrainuniv.edu.iq

1. Introduction

Noise, the unwanted and damaging sound levels [1] has dramatically increased due to the recent increase of global technological developments including cars, factories, aircraft, and others that cause noise stress which is one of the leading global causes of environmental stress [2]. form of stress causes ultra-structural changes in the fascial area of the adrenal cortex and catecholamine granule of the adrenal medulla, which is followed by a rise in levels of plasma corticosterone. This fact caused the corticosterone level to be a mark of stress [1]. The noise stress affects our bodies depending on the exposure duration. Long-term noise stress could be more harmful than acute since it affects many body systems and causes many health problems such as respiratory damage poor diet, and sleep insomnia [3]. These effects are mediated through the action of stress on the immune system [4]. Immunoglobulins (Igs) are proteins that are important to the immune system; they bind to foreign substances, including germs, and help to eliminate them [5]. In response to the antigenic stimulation of naive B cells, they are synthesized and secreted by plasma cells [6]. There are five isotypes of Ig which are IgM, IgG, IgA, IgE, and IgD [7]. The most common isotype in

the body is IgG, so it is the major Ig in serum (systemic immunity) and it is the major Ig found in extravascular spaces. Of all Igs, it has the longest serum half-life. Additionally, IgG antibodies directly support an immune response by neutralizing pathogens and viruses, which are then engulfed by phagocytosis and eliminated [8]. Some researchers showed that exposure to noise increases the levels of IgG and IgM (Dhabhar, 2001) but how does it differ between acute and chronic exposure? And which gender could be affected by noise exposure more than the other? To answer these questions, this study aimed to demonstrate the effect of 90 dB noise stress during 4 hours daily for 1, 8, 14, 21 and 30 consecutive days of stress on the immunological status of adult mice male and female.

2. Materials and Methods

Balb/C mice, aged between 6-8 weeks and weighted 20-28 g were used. They were provided by the Iraqi Center for Cancer Research and Medical Genetic-Ministry of Higher Education and Scientific Research. Animals were housed in the cages (5 males or females in each cage) under standard and conventional conditions, stable temperature and 12 h light-dark cycles, they were also provided with mice pellets and tap water *ad leptum*. After 2-week adaptation, fifty Mice were randomly assigned and prepared for noise stress exposure, in contrast, 10 mice (5 males and 5 males) were left without being stressed, this group was used as a control group (C). Broadband white noise (20-20,000) Hz at (90 dB) intensity [1], was applied by application of an android (frequency sound generator and sound level meter) to measure sound intensity. The phone was connected by bluetooth to the multimedia speaker (BK_868) which was fixed at 30 cm over the animal cages. During the exposure period to noise stress, these animals are kept in the stress room to prevent other unnecessary stress on the animals.

Mice exposed to stress were divided into 5 groups (n=10, 5 males and 5 females): Group 1 (G1) mice were exposed to 90 dB noise intensity for 4 hours, and the mice were sacrificed on the same day; Group 2 (G2) mice were exposed to 90 dB noise intensity 4 hours daily during 8 days, then the mice were sacrificed at the 9th day; Group 3 (G3) mice were exposed to 90 dB noise intensity 4 hours daily during 14 days, then the mice were sacrificed at the 15th day. Chronic noise stress groups; Group 4 (G4) mice were exposed to 90 dB noise intensity for 4 hours daily for 21 days, then the mice were sacrificed on the 22nd day. Group 5 (G5) mice were exposed to 90 dB noise intensity for 4 hours daily for 28 days, and the mice were sacrificed on the 29th day. After the end of the experiment, the mice were weighed and sacrificed by neck dislocation. The blood samples were collected from the eye of each mouse in two tubes; a sterile gel tube that allowed clotting to separate the serum and a sterile tube with anticoagulant that was used for the hematological test by Diagon-Cell 60 (D-Cell 60) Hematology Analyzer. Serum was separated by centrifugation of the blood sample in gel tubes at (10000) rpm for (10-15) mins, and kept at -20C°. The serum was used for the determination of glucocorticoids, IgG and IgM levels. Levels of glucocorticoids were determined by Cobas e-411 analyzer device.

The kit that used in this study was from Rosh company (Germany). Whereas IgG and IgM levels were measured by a quantitative turbid metric assay using linear chemicals and immunoassay fully automatic device Linear (INC) from linear company/Spanish. Results were given as mean±SD. For multiple comparisons and using Statview version 5.0, data were analyzed using one-way analysis of variance (0ne- way ANOVA), which is followed by Fisher's test, when $p<0.05$ differences were deemed significant.

3. Results and Discussion

Table 1 shows a high significant ($P<0.01$) decrease in WBC count in 1, 8, and 14-day stress group (3.14 ± 0.28, 4.82 ± 0.62, and 4.65 ± 1.27 x109/L, respectively in males and 3.24±0.28, 4.92 ± 0.62, and 4.85±1.82 x109/L, respectively in females) compared to control group (5.64±0.45 x109/L in males and 5.75 ± 0.44 x109/L in females). Interestingly, the total WBC was highly significant ($P<0.01$) in the 21-day stress group, while there was no significant difference in the 30-day stress group (6.34 ± 0.26 and 5.63 ± 0.44 x109/L, respectively in males and 6.74 ± 0.36 and 5.93 ± 0.44, in females respectively). Table 2 shows a highly significant increase ($P<0.01$) in glucocorticoids level in the stress groups during 1, 8, 14, 21, and 30-day (3.81 ± 1.31, 4.22 ± 1.315, 9.57 ± 0.907, 13.448 ± 1.606, 19.48 ± 3.348 nmol/L) respectively in males, and (13.072 ± 1.79, 22.304 ± 1.38, 16.994 ± 0.50, 45.782 ± 1.50, 39.462 ± 1.91

nmol/L), respectively in females in compared with control groups (3.378 ± 1.16 in males and 3.984 ±1.48 in females). These data were significantly higher (P<0.01) in females compared with the male groups. This increase could reach to about 3 or 4 times (Table 2). On the other hand, (Table 3) shows the mean comparison of IgG mg/dl, and IgM mg/dl between different groups by comparing the control groups with other groups that are under the stress of noise during (1, 8, 14, 21, and 30) days, as well by comparing male with female groups. IgG levels significantly increased in the noise stress groups; 1, 8, 14, 21, and 30 days compared to control groups (99.8 ± 10.7 in males and 105.3 ± 17.9 mg/ml in females). These levels significantly increased in noise stress group; 8, 14, and 21 days (213.4±20.4, 213.9±36.3, and 219.6 ±2 5.9 mg/ml, respectively in males and 184.3 ± 15.0, 196.5 ± 30.7, and 221.4 ± 26.3 respectively in females) compared to day 1 (181.1 ± 37.7 in males and174.3 ± 26.6 mg/ml in females), after that return back to decrease significantly in 30-days stress group (177.4 ± 13.3 in males and 178.4 ± 8.1 mg/ml in females) compared to the rest of noise stress groups. Essentially, the same differences are observed in IgM levels at the same time. IgM levels significantly increased in noise stress groups; 1, 8, 14, 21, and 30 days compared to controls (93.9 ± 6.1 in males and 81.2 ±7.7 mg/ml in females) except in the 30-day group of females (85.4 ±5.9 mg/ml) which did not have a significant difference compared to the control group. These levels increased significantly in noise stress group; 8, 14, and 21 days (105.6 ±11.7, 109.1 ±16.1, and 117.1 ±5.2 mg/ml, respectively in males and 98.9 ±10.6, 113.4 ±10.8, and 119.8±9.5, respectively in female) compared to day 1 (106.6 ± 9.8 in male and 95.4 ± 5.8 in females), after that back to decrease significantly in 30-day stress group (101.6 ± 9.4 in males and 85.4±5.9 mg/ml in females) compared to the rest noise stress groups. There were no significant differences in IgG and IgM levels between males and females except in 8- and 14-day noise stress groups. In these groups, levels of IgG and IgM were significantly higher (P<0.01) in males when compared with the females.

Stress has been reported to produce profound effects on the immune system and physiological parameters of the body. The immune system once assumed to be autonomous, is now known to respond to signals from many of the body's systems, especially the endocrine and nervous systems. Stress influences hormones that bind to particular receptors on the membrane or in the cytoplasm of immune system cells, including multiple cells that participate in antibody production [9]. Moreover, glucocorticoids are utilized as a marker for stress activation in both humans and animals since it is well known that stress increases the activity of the hypothalamic-pituitary-adrenal axis (HPA axis) and causes the adrenal cortex to secrete more glucocorticoids which agreed with our results that glucocorticoids increased after acute and chronic noise stress exposed [10,11]. However, sex differences have been reported as well in HPA axis activation in many studies. Zavala reported that female rats enhanced hypothalamus activation after exposure to acute and chronic stress compared to males [12]. Iwasaki-Sekino mentioned the impact of the estrous cycle on HPA axis activity. When exposed to an acute stressor, the proestrus phase in female rats is marked by an increase in the progesterone serum level, and estradiol shows more activation in the paraventricular nucleus of the hypothalamus than do male rats [13]. This could explain the increased levels of glucocorticoids in females more than in males in all studied time points.

Our results showed a significant decrease in total WBC in mice exposed to stress of noise during 1, 8, and 14 days which were increased in 21-day stress groups to be similar to control groups in 28-day stress groups in both males and females. Our results were in agreement with the observations of [14], who reported a significant decrease in total leukocyte count in male rats exposed to 100 dB noise stress for 4 hours. They explained the indicated leukopenia by the excessive release of glucocorticoids in stress and changes in other factors like neurotransmitters, pituitary peptides, and adrenal hormones which could be the physiological mediators caused this change observed [14] While [15], concluded that the decrease in blood leukocytes number may be interpreted in two possible ways, it could reflect large scale destruction of circulatory leukocytes [15]. Alternatively, it can be an indication that leukocytes are moving from the blood to different body organs Additionally, it has been demonstrated that the release of glucocorticoids alters a variety of immunological characteristics, such as the location of immune cells when cell death

is not present.[16]. It is also possible to hypothesize that stress-related conditions that activate the hypothalamic-pituitary-adrenal axis (HPA) lead to a decrease in the number of leukocytes in the bloodstream; these conditions frequently happen in the later stages of the stress response, during prolonged acute stress (lasting for hours), or during extremely stressful psychological or physical situations. to prepare for immune challenges that may be imposed by the actions of the stressor, activation of the HPA axis causes the release of glucocorticoid hormones, which cause leukocytes to exit the blood and settle at the skin, mucosal lining of the gastrointestinal, urinary genital tracts, lung, liver, and lymph nodes. As a result of this redistribution of leukocytes, the number of blood leukocytes decreases, the effect is most prominent in monocytes, T and B lymphocytes [15],[16]. and this may be the cause of increased serum levels of IgG and IgM despite the WBCs count decrease as was found in this study in 1,8,14, and 21 noise stress days groups. There is substantial evidence indicating that long-term stress and anxiety promote the production of proinflammatory cytokines, such as IL-6 which is associated with the lower levels of B and T lymphocytes. This may lead to a decrement in the level of immunoglobulin, IgG, and IgM, as there is an increase in the duration of stress [17] which agrees with our results in the 30-noise stress exposure day group. On the other hand, adrenergic pathways are the primary means of communication between the immune system and the neurological system. In comparison to innate immunity, their involvement in modulating adaptive immunity has garnered more attention [18]. however consistent data suggest that adrenergic processes also play a crucial role in immune cells [19] which could cause a decrease in the WBC migration to the organs and raise their numbers in the blood flow in addition to a decrease in IgG and IgM levels. The evidence may explain the difference between immunoglobulin levels in both the short and long term which were found in this study.

Table 1. Effect of Noise Stress on White Blood Cell (WBC) Indices in Male Mice

Groups		White Blood Cell Indices (mean ± SD)x10⁹/L				P value
		Total WBC	Lymphocyte	Monocyte	Granulocyte	
Control		5.64±0.45a	1.69±0.21a	0.56±0.01a	3.39±0.12a	≤ 0.01
Acute Stress	1 day	3.14±0.28d	0.94±0.01d	0.31±0.01d	1.89±0.01d	
	8 days	4.82±0.61c	1.45±0.32c	0.48±0.03c	2.89±0.11c	
	14 days	4.65±0.26c	1.39±0.62c	0.47±0.02c	2.79±1.13c	
Chronic Stress	21 days	6.34±0.26b	1.90±0.71b	0.63±0.01b	3.80±0.98b	
	30 days	5.63±0.44a	1.69±0.51a	0.56±0.02a	3.38±1.21a	
LSD		1.9	1.0	0.4	0.6	

Table 2. Effect of Noise Stress on White Blood Cell (WBC) Indices in Female Mice

Groups		White Blood Cell Indices (mean ± SD)x10⁹/L				P value
		Total WBC	Lymphocyte	Monocyte	Granulocyte	
Control		5.44±0.44a	1.51±0.60a	0.59±0.01a	3.34±1.8a	≤ 0.01
Acute Stress	1 day	3.44±0.18d	0.99±0.23d	0.36±0.01d	2.09±0.87d	
	8 days	4.32±0.32c	1.40±0.42c	0.49±0.17c	2.43±0.93c	
	14 days	4.15±١.82c	1.34±0.72c	0.45±0.13c	2.36±0.83c	
Chronic Stress	21 days	6.24±0.36b	1.99±0.81b	0.66±0.23b	3.59±0.78b	
	30 days	5.33±0.24a	1.59±0.11a	0.60±0.23a	3.14±1.32a	
LSD		2.0	1.5	0.3	0.6	

Table 3. Effect of Noise Stress on the level of Cortisol Hormone in Male and Female Mice

Groups		Cortisol hormone nmol/L (mean ± SD)		P value
		Male	Female	
Control		3.378±1.167 a	3.984±1.484 a	≤ 0.01
Acute Stress	1 day	3.810±1.310 b	13.072±1.792 b*	
	8 days	4.220±1.315 b	22.304±1.380 c*	
	14 days	9.570±0.907 c	16.994±0.504 d*	
Chronic Stress	21 days	13.448±1.606 a	45.782±1.505 f*	
	30 days	19.480±3.348 d	39.462±1.912 e*	
LSD		4.100	9.00	

Table 4. Effect of Noise Stress on Immunoglobulins in the male and Female Mice

Groups		IgG mg/dl (mean ± SD)		IgM mg/dl (mean ± SD)		P value
		Male	Female	Male	Female	
Control		99.8±10.7a	105.3±17.9a	93.9±6.1a	81.2±7.7a	≤ 0.01
Acute Stress	1 day	181.1±37.7c	174.3±26.6b	106.6±9.8c	95.4±5.8b	
	8 days	213.4±20.4d*	184.3±15.0c	105.6±11.7c	98.9±10.6b	
	14 days	213.9±36.3d*	196.5±30.7d	109.1±16.1c	113.4±10.8c	
Chronic Stress	21 days	219.6±25.9d	221.4±26.3e	117.1±5.2d	119.8±9.5c	
	30 days	177.4±13.3b	178.4±8.1b	101.6±9.4b	85.4±5.9a	
LSD		76	30	30	25	

- The difference in the letters means there is a significant difference among means in columns ($p \leq 0.01$), while the similar letters denote a non-significant difference among means in columns ($p \geq 0.05$). * it means there is a significant difference between females and males.

4. Conclusions

The results in this work showed that the exposure to noise stress increased glucocorticoid levels by activation of the HPA axis which could affect the distribution of WBCs in the organs leading to a decrease of its count in the blood with an increase of IgG and IgM levels. These changes differed in the acute noise exposure than the chronic exposure which returned to normal in the 30-day exposure. On the other hand, these changes differed in females compared to males which might be because of the enhancement of hypothalamus activation in females after exposure to acute and chronic stress in comparison to males. Thus, the current study recommends further researches that extend to more than 30 exposed days to know whether this value continued to drop or not which could be very harmful to the body. Moreover, these results can lead to many perspectives such as the determination of neurotransmitters and hormones to prove the theory discussed above.

Acknowledgments: The authors are grateful to the Department of Biology College of Science at Al-Mustansiriyah University for supporting this project.

Funding: No funding is received for this work.

Conflicts of Interest: There are no conflicts of interest.

References

[1] Salehpour F.; Mahmoudi J.; Eyvazzadeh N.; "Effects of acute and chronic noise stress on depressive-and anxiety-like behaviors in mice". JECNS, 5(1): 1-6, 2018.

[2] Barzegar M.; Sajjadi F. S.; Talaei S. A.; Hamidi G.; Salami M.; "Prenatal exposure to noise stress: anxiety, impaired spatial memory, and deteriorated hippocampal plasticity in postnatal life". Hippocampus, 25(2): 187-196, 2015.

[3] Khanam S.; "Impact of stress on physiology of endocrine system and on immune system: A Review". Inter. J. Diabetes Endocrinol., 2(3): 40-42, 2017.
[4] Vitlic A.; Lord J.M.; Phillips A.C.S.; "Ageing and their influence on functional, cellular and molecular aspects of the immune system". Age, 36(3): 1169-1185, 2014.
[5] Khan F.A.; "Biotechnology in Medical Sciences". CRC Press: 328-341, 2014.
[6] Ball, G.F.; "Vitamins: their role in the human body". John Wiley & Sons, 2008.
[7] Stavnezer J.; Guikema J.E.; Schrader C.E.; "Mechanism and regulation of class switch recombination". Annu. Rev. Immunol., 26(261), 2008.
[8] Janeway J.C.A.; Travers P.; Walport M.; Shlomchik M.J.; "The distribution and functions of immunoglobulin isotypes". In Immunobiology: The Immune System in Health and Disease. 5th edition. Garland Science., 2001.
[9] Hitti, M.; Chang L.; "Chronic fatigue syndrome linked to hormone". J. Clin. Endocrinol. Metab., 22: 326-30, 2008.
[10] Podsevatkin V.G.; Kiriukhina S.V.; Podsevatkin D.V.; Podsevatkina S.V.; Blinov D.S.; "Dynamics of the behavioral response and cortisole level caused by the combined action of mexidole, diazepam, thymogen, and hyperbaric oxygenation in mice under immobilization stress conditions". Eksp. Klin. Farmakol., 71(1): 22-25, 2008.
[11] Zhang J.; "Basic neural units of the brain: neurons, synapses and action potential". arXiv preprint arXiv:1906.01703, 2019.
[12] Zavala J.K.; Fernandez A.A.; Gosselink K.L.; "Female responses to acute and repeated restraint stress differ from those in males". Physiol. behav., 104(2): 215- 221, 2011.
[13] Iwasaki-Sekino, A.; Mano-Otagiri, A.; Ohata, H.; Yamauchi, N.; Shibasaki, T.; "Gender differences in corticotropin and corticosterone secretion and corticotropinreleasing factor mRNA expression in the paraventricular nucleus of the hypothalamus and the central nucleus of the amygdala in response to foot shock stress or psychological stress in rats". Psychoneuroendocrinology, 34(2): 226-237, 2009.
[14] Archana, R.; Namasivayam, A.; "The effect of acute noise stress on neutrophil functions". Ind. J. Physiol. Pharmacol., 43(4), 1999.
[15] Dhabhar F.S.; "Stress-induced augmentation of immune function—the role of stress hormones, leukocyte trafficking, and cytokines". Brain, Behav. Immun., 16(6): 785-798, 2002.
[16] Dhabhar F.S.; "Bidirectional effects of stress and glucocorticoid hormones on immune function: possible explanations for paradoxical observations". Psychoneuroimmunology, 2001.
[17] Segerstrom, S.C.; Miller, G.E.; "Psychological stress and the human immune system: a meta-analytic study of 30 years of inquiry". Psychological Bulletin, 130(4): 601, 2004.
[18] Marino, F.; Cosentino, M.; "Adrenergic modulation of immune cells: an update". Amino Acids, 45(1): 55-71, 2013.
[19] Scanzano, A.; Cosentino, M.; "Adrenergic regulation of innate immunity: a review". Front. Pharma., 6:171, 2015.