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The Effects of Sound Pollution on the Serum Levels of Corticosterone and Other Hematological Parameters in Male Rats

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ABSTRACT

BACKGROUND AND OBJECTIVE: Sound pollution is one of the main causes of stress that could lead to several physiological and mental changes in humans. This study aimed to investigate the effects of sound pollution on the serum levels of corticosterone and other hematological parameters in male rats.

METHODS: In this experimental study, 60 male Wistar rats were randomly divided into two groups of short-term experiment (one-day exposure) and long-term experiment (30-day exposure). Each group consisted of one control group and three experimental groups with 4, 8 and 12 hours of exposure to noise pollution. According to a pre-designated schedule, the animals were exposed to 100 dB sound pressure, and changes in the serum levels of corticosterone and hematological parameters were recorded.

FINDINGS: In this study, serum levels of corticosterone increased in the long-term exposure group (8 hours: 5.36±3.19 ng/dl, 12 hours: 4.88±1.76 ng/dl) compared to the control group (1.08±0.53 ng/dl) ($p<0.01$ and $p<0.001$, respectively). Number of red blood cells also increased in the long-term exposure group (8 hours: 7.71±0.33 mm^3, 12 hours: 8.22±0.69 mm^3) compared to the control group (6.24±0.16 mm^3) ($p<0.05$ and $p<0.01$, respectively). In addition, there was an increase in hemoglobin concentration in the long-term exposure group (8 hours: 16.66±0.68 gr/dl, 12 hours: 16.60±0.81gr/dl) compared to the control group (14.15±0.66 gr/dl) ($p<0.05$). Moreover, there was a significant increase in the hematocrit in the two groups of 8 hours (40.57±3.28) and 12 hours (40.66±1.76) of long-term exposure compared to the control group (35.69±3.13) ($p<0.05$).

CONCLUSION: According to the results of this study, long-term exposure to noise pollution could increase the number of red blood cells and other hematological parameters by raising the serum levels of corticosterone.

KEY WORDS: Sound Pollution, Corticosterone, Complete Blood Cell Count, Rat

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Introduction

Sound pollution is one of the most severe consequences of modern life in industrial communities. Rapid development of urban life and continuous expansion of industries on the outskirts of cities have endangered the peaceful and quiet living of humans, causing numerous environmental issues. With the dramatic growth of noise pollution sources, humans are constantly exposed to this health hazard; at home, at work or while walking in the streets.Noise is considered as a major stress-causing agent for the body, and this type of pollution could cross through any obstacles

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affecting a population involuntarily (1). For ages, people have been aware of the harmful effects caused by sound pollution, attempting to devise new strategies everyday as to battle this environmental inconvenience (2, 3). Long-term or short-term exposure to sound pollution disrupts many physiological functions in different systems of the body in humans and animals, including the nervous system, endocrine glands and cardiovascular system (4-6). In addition, consecutive or abrupt subjection to loud noises may lead to certain responses in the body; such examples are changes in the heart rate, blood pressure, respiratory rate, gastrointestinal motility, secretion of different hormones from the endocrine glands and learning and memory defects (7,8). These reactions may appear as alerts or stress responses, and in terms of physiological features, they are not easily distinguishable from other body reactions to stimuli such as fear or anger. Short-term exposure to noise often yields temporary effects (9, 10); however, these effects were observed to become permanent in case of long-term exposure to noise pollution in laboratory animals, most of which involve the cardiovascular system (11). Exposure to any noise higher than 90 dB is normally considered as a source of stress (12). Acute and chronic exposure to noise leads to the production of free radicals, such as super oxidase, catalase and glutathione peroxidase (13).

Moreover, constant exposure to 100 decibels of sound pressure could result in liver toxicity, decreased antioxidant activity and increased blood lipid peroxidation parameters (14). On the other hand, continuous exposure to low-frequency noise pollution with intensity of 33-52 dB could lead to the hypersecretion of cortisol during the early hours of night, which could severely disrupt the daily rhythm of cortisol secretion (15, 16). Furthermore, low sound levels during sleep might increase cortisol secretion. This occurs due to the close contact of neurons in cerebral cortex sub-regions and parts of the auditory system. The neuroplasticity of these cells affects the function of hypothalamic-pituitary-adrenal axis (17). In Iran, with the focus of researchers on air pollution and its side effects, there is insufficient information on the biological effects of noise pollution, while in other countries, several studies have been conducted on the cardiovascular effects caused by sound pollution in recent years (18-23).Cardiovascular diseases are among the most important health issues in modern, industrial societies. Although provision of patient treatment imposes a heavy cost on the health care system of every country, cardiovascular diseases account for a noticeable mortality rate every year. Therefore, identification of the effects of noise pollution on the physiological parameters of living organisms is of paramount importance in order to reduce the adverse effects on the auditory system. The present study aimed to investigate the effects of short-term and long-term exposure to 100 dB noise pollution on the secretion of serum corticosterone and other hematological parameters in male rats.

Methods

Animals and Experimental Groups: In this experimental study, 60 adult, male Wistar rats (weight: 250-300 g) were purchased from the Department of Animal Breeding at Razi Vaccine and Serum Research Institute and transferred to the animal house. The rats were kept in special cages under controlled conditions within the light cycle of 12 hours of light and 12 hours of darkness at the temperature of 25±2°C.During the study, the animals had access to sufficient food and water, and all the experiments were performed in accordance with the guidelines of working with laboratory animals (American National Institutes of Health, Publication No. 80-23, Revised 1996). The study protocol was approved by the Ethics Committee of Sabzevar University of Medical Sciences, Iran. When the animals were accustomed to their condition after one week, they were randomly divided into two experimental groups of long-term exposure and short-term exposure to noise pollution. The short-term exposure group consisted of four subgroups: three groups receiving exposure for 4, 8 and 12 hours (5 rats in each subgroup), and one control group. For short-term exposure, the animals were placed in a room and subjected to 100 dB noise only once for 4, 8 and 12 hours.During this experiment, the control subjects were placed in a separate room without any exposure. The long-term exposure group consisted of four subgroups: three experimental groups receiving noise exposure for 4, 8 and 12 hours (10 rats in each group), and one control group. In order to investigate the effects of long-term exposure, the animals were placed in a room for 30 days and subjected to 100 dB noise for

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4, 8 and 12 hours per day (14). The control subjects were kept in a separate room without any noise pollution. In this study, exposure started at 6 pm for the experimental groups.

Implementation of Noise Pollution: In this study, the desired noise pollution was created using computer software, recorded on a cassette tape and played with a Sony stereo player (made in Japan).In order to improve the sound volume, powerful amplifiers and speakers were installed in the room. During the experiment, 100 dB sounds were regularly monitored in the four corners of the room using calibrated sound level meters (PCE Instruments, Ver. 318). Four rats were placed in each cage, and the cages were located at a specific distance from the stereo player. The animals were exposed to the noise at different intervals of 4, 8 and 12 hours.

Blood Samples: In this study, all the animals were weighed before the experiment. At the end of the experiments, the rats were anesthetized, and 5 ml of blood was collected from the dorsal aorta of each animal with a surgery. One ml of the obtained blood samples was transferred into heparin microtubes in order to perform complete blood cell count (CBC). In addition, serum was extracted from the rest of the blood, moved into capped microtubes and preserved at -20°C until the initiation of Radioimmunoassay tests (RIA) for measuring corticosterone concentration.

Complete Blood Cell Count: For complete blood cell count, one ml of fresh blood was moved to the laboratory and analyzed using Sysmex Amercia, Inc. device. Different blood parameters including the number of red blood cells, white blood cell count, platelet count, hemoglobin concentration, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean cell hemoglobin concentration (MCHC) were measured as well.

Measurement of Serum Corticosterone Levels using the RIA Technique: Serum concentrations of corticosterone were measured using solid-phase radioimmunoassay kit (DRG Inc. USA) in accordance with the protocols of the manufacturer. In this model, marked corticosterone with I^{125} competes against the corticosterone of samples for attaching to antibodies. After incubation, the tubes were flushed so that the free corticosterone would disconnect from the attached corticosterone to antibodies, and the competition would be terminated. Measurement of radioactivity using a gamma counter device results in a figure, which is the measure value of corticosterone in serum samples based on the standard curve.

Statistical Analysis: Data obtained from the measurement of different parameters were analyzed using SPSS V.16, one-way ANOVA and post-hoc Tukey's test, and $p<0.05$ was considered as significant.

Results

Concentration of Serum Corticosterone in Short-term and Long-term Exposure Groups: In this study, short-term exposure to noise was observed to raise the serum levels of corticosterone in the experimental groups, especially in the subjects receiving 8 hours of noise pollution, compared to the control group; however, this increase was not considered to be statistically significant (table 1).

On the other hand, long-term exposure could significantly increase the concentration of serum corticosterone in the groups receiving 8 and 12 hours of noise pollution compared to the control group ($p<0.01$ and $p<0.001$, respectively). In addition, no significant increase was observed in the experimental group subjected to 4 hours of noise pollution (table 1). **Effects of Noise Pollution on Hematological Parameters of the Animals in the Long-term Exposure Group:** According to the results of this study, long-term exposure to noise pollution could significantly increase the number of red blood cells ($p<0.05$, $p<0.01$), hematocrit ($p<0.05$) and hemoglobin concentration ($p<0.05$) in the groups subjected to exposure for 4, 8 and 12 hours for 30 days compared to the control group.

However, this difference was not significant in the animals receiving 4 hours of exposure during 30 days. Moreover, the results of this study indicated that exposure to noise pollution had no significant effects on MCV, MCHC and the total number of white blood cells (table 2). It is also noteworthy that the short-term effects of noise exposure on the hematological parameters of the animals were not investigated in this study

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Table 1. Mean Concentration of Serum Corticosterone (ng/dl) in Short-term and Long-term Exposure Groups

Groups / Type of Exposure	Control	4 Hours a Day	8 Hours a Day	12 Hours Day
Short-term	2.04±0.18	2.74±1.54	4.23±2.25	2.85±1.43
Long-term	1.08±0.53	3.27±3.08	**5.36±3.19	***4.88±1.76

**p<0.01, ***p<0.001, significant difference with the control group

Table 2. Mean of Blood Cell Parameters in Different Long-term Exposure Groups

Groups / Hematological Parameters	Control	4 Hours a Day	8 Hours a Day	12 Hours a Day
Number of Blood Cells per Cubic Millimeter (10^6)	6.24±0.16	6.78± 0.94	*7.71±0.33	**8.22±0.69
Hemoglobin Level (g/dl)	14.15±0.66	13.23±2.1	*16.66±0.68	*16.60±0.81
Hematocrit (%)	35.69±3.13	35.01±5.34	*40.57±3.28	*40.66±1.76
Mean MCV (fL)	49.98±4.21	52.08±1.97	52.93±6.45	48.62±2.61
Mean MCH (pg)	19.52±0.86	19.45±3.85	20.38±1.31	19.11±2.12
Mean MCHC (g/dL)	39.38±4.82	37.34±0.44	38.87±4.17	39.24±3.01
Number of White Blood Cells per Cubic Millimeter (10^3)	8±1.34	10.03±6.23	10.50±6.23	9.65±2.73

**p<0.01, ***p <0.001, significant difference with the control group

Discussion

According to the results of this study, long-term exposure to noise pollution (30 days) could significantly increase the serum levels of corticosterone in male rats. Moreover, long-term noise exposure could lead to an increase in the number of red blood cells, hematocrit and serum concentration of hemoglobin. Several studies have reported that subjection to different physical and mental stressors could increase the secretion of stress hormones, such as cortisol, corticosterone, adrenaline and noradrenaline (24, 25). In addition, long-term exposure to sound pollution is likely to raise the cortical volume of the adrenal gland, which will lead to the increased activity of this gland (26).

Loud and sudden noises are physical stress-causing factors, which could activate the hypothalamic-pituitary-neural axis of the adrenal gland cortex and increase the secretion of stress hormones from this gland (16, 24). Increased number of red blood cells, hematocrit and hemoglobin concentration following the long-term exposure to noise pollution, as observed in this study, is probably indicative of enhanced hematopoiesis in the bone marrow, which occurs due to noise stress. Nevertheless, the physical characteristics of red blood cells remained unchanged; in other words, produced red blood cells were natural in terms of shape, mass and hemoglobin content. In accordance with this finding, long-term and short-term exposure to disturbing, loud noises were observed to cause certain changes in the hematological and biochemical features of blood, including liver toxicity, decreased antioxidant parameters and increased enzymatic lipid peroxidation in humans and animals (27-30).

Constant exposure to noise pollution could increase the concentration of serum corticosterone by activating the hypothalamic-pituitary-adrenal axis of the adrenal gland cortex (31); consequently, production of red blood cells may rise in the bone marrow. According to the literature, continuous increase in the serum concentrations of corticosterone and cortisol could frequently lead to polycythemia, while decreased secretion from adrenal gland could result in the occurrence of anemia (32). It is presumed that corticosterone increases the production of red blood cells by enhancing the growth and cell division in the bone

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marrow; on the other hand, this hormone could influence the expression of the genes responsible for hemoglobin synthesis, which results in increased hematocrit (32-35). Chronic increase of corticosteroids may incapacitate the immune system; presence of lymphocytopenia and eosinopenia could be a proper diagnostic criterion regarding the excessive production of corticosterone by the adrenal gland. In addition, high levels of cortisol are associated with the increased number of neutrophils (36, 37). The findings of the current study are inconsistent with the results obtained by Archana et al. and Weisse et al., which indicated that long-term exposure to noise pollution could decrease the number of white blood cells and cause changes in the nervous system in humans (27,30,37). This difference could be due to the fact that the aforementioned studies were conducted based on the differential evaluation of white blood cells, while in the present study, we calculated the total white blood cell count only. Long-term exposure to noise pollution is likely to increase the secretion and serum concentration of stress hormones, such as corticosterone and cortisol, by stimulating the activity of the hypothalamic-pituitary-adrenal axis. It is also noteworthy that a temporary increase in the level of corticosterone could contribute to the better adaptation of an individual with stressful situations, while constant elevation in the concentration of this hormone could have destructive effects on the body; such examples are weakened immune system and vulnerability of the body to different pathogens (36, 38). In conclusion, the findings of the current study indicated that long-term exposure to irritating noises could cause a significant increase in the secretion and serum concentration of corticosterone. Furthermore, chronic exposure to noise pollutions may raise the number of red blood cells, affecting other hematological parameters as well. However, it has no significant effects on the total white blood cell count and other indices such as MCH, MCV and MCHC.

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