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# Effect of noise exposure (85 dB) on testicular adrenocortical steroidogenic key enzymes, acid and alkaline phosphatase activities of sex organs in mature albino rats

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**Abstract:** Changes in the activities of  $\Delta^5$ - $3\beta$ -hydroxysteroid dehydrogenase (HSD) in testis and adrenal gland,  $17\beta$ -hydroxysteroid dehydrogenase in testis, acid and alkaline phosphatase in testis, prostate and seminal vesicle were observed in noise exposed mature rats at the intensity of 85 dB for 8 h/day for 45 days. The results indicated that noise exposed group showed a significant diminution in the activities of androgenic key enzymes  $\Delta^5$ - $3\beta$  and  $17\beta$ -HSD, acid phosphatase in testis, prostate and seminal vesicle. There was a significant elevation in the activities of adrenal  $\Delta^5$ - $3\beta$ -HSD, alkaline phosphatase in testis and other accessory sex organ in noise exposed group. Gonadosomatic, prostatosomatic and seminal vesiculo-somatic indexes were decreased significantly in noise exposed group. Therefore, it is evident that noise exposure at 85dB exerts a deleterious effect on testicular and adrenocortical activities.

**Key words:** noise; hydroxysteroid dehydrogenase; acid phosphatase; alkaline phosphatase; sex organs; adrenal cortex

## Introduction

For last two decades, a wave of the experimental consciousness is being developed in developing countries including India. Modern technology have been created environmental pollutant, of which noise is considered as the worst enemy of human health and a curse of our society. Noise is also an occupational hazard in industrial workers. Moreover, general individuals are frequently exposed in road traffic noise. From literature survey it has been revealed that noise exposure is associated with hypertension (Grandjean, 1988), disturbance of sleep (Grandjean, 1988), deafness (Grandjean, 1988; Chhatwal, 1995) heart rate acceleration (Grandjean, 1988) and premature delivery (Bragden, 1970) but there is no report about the effects of noise on male reproduction and adrenocortical activities. As noise intensity of most of the industries is within the range of 60 dB to 110 dB (Chhatwal, 1995) and of road traffic noise is within 77-94 dB (Chhatwal, 1995), so here we select 85 dB, the average intensity of sound of these above sources. Therefore the present study was undertaken to find out the deleterious effect of noise on testicular and adrenocortical activities considering steroidogenic key enzymes in these organs and acid as well as alkaline phosphatase activities in sex organs which are sensitive functional indicators of reproductivity status of the animal.

## 1 Material and methods

### 1.1 Animal and noise exposure

16 adult male albino Wister rats having the age of 60 days and body weight  $120 \pm 5$ g were used in this experiment. They were acclimatized to the laboratory conditions by providing them with standard diet and water *ad libitum*. Animals were placed in normal photoperiod (12L:12D) with the animal house temperature set around  $28^\circ \pm 1^\circ\text{C}$ . Animals were divided into 2 equal groups. Animals of one group were exposed to 85 dB sound for 8 h (8 a. m.—4 p. m.) per day for 45 days and considered as experimental group. Rest part of the day, these experimental rats were placed in domestic noise at the intensity level of 15–30 dB. Remaining group was considered as control and these rats were exposed to domestic noise at day time. At night, both control and experimental groups were placed in calm and quiet environment having the sound intensity of 5–10 dB. On 46th day of experiment, animals were sacrificed, body weights were noted. Testes, prostate, seminal vesicle and adrenals were dissected out and their weight were noted.

Chemicals: NAD, DHEA, testosterone, glycerol, tereasodium pyrophosphate, tris chloride,

acetic acid, cobalt chloride, sodium fluoride, p-nitro-phenolphosphate, potassium phosphate, propylene glycol, crystalline BSA and sodium hydroxide used in this experiment were purchased from BDH or Sigma Chemical Company.

### 1.2 Assay of testicular $\Delta^5$ - $3\beta$ and $17\beta$ -HSD

One testis from each animal was used to study the activities of  $\Delta^5$ - $3\beta$ -HSD according to the method of Talalay (Talalay, 1962). Testicular tissue was homogenized at a tissue concentration of 100 mg/ml of homogenising mixture which contains 20% spectroscopic grade glycerol, 5 mmol/L potassium phosphate and 1 mmol/L EDTA. This homogenizing mixture was centrifuged at 10000 g for 30 min at 4°C. The supernatant (1ml) was mixed with 100 $\mu$ mol sodium pyrophosphate buffer (pH 8–9) and 30 $\mu$ g of dehydroepiandrosterone (DHEA) making the incubation mixture a total of 3 ml. Enzyme activity was measured after addition of 0.5 $\mu$ mol of NAD to the tissue supernatant mixture in a spectrophotometer at 340 nm against a blank (without NAD<sup>+</sup>). 1 unit of enzyme activity is the amount causing a change in absorbance of 0.001/min at 340 nm.

Testicular  $17\beta$ -HSD activity was measured by the method described by Jarabak *et al.* (Jarabak, 1962). Testicular tissue containing  $\Delta^5$  -  $3\beta$ -HSD was homogenized at a tissue concentration of 100 mg/ml of this mixture. After centrifugation at 10000 g for 30 min at 4°C of this homogenized tissue preparation, supernatant was collected. 1 ml of this supernatant was mixed with 440 $\mu$ mol of sodium pyrophosphate buffer (pH 10.2), 25 mg crystalline BSA and 0.3 $\mu$ mol testosterone making the incubation mixture of a total 3 ml. Enzyme activity was measured after addition of 1.1 $\mu$ mol NADP to the tissue supernatant mixture in a spectrophotometer at 340 nm against a blank (without NADP). One unit of enzyme activity is equivalent to a change in absorbency of 0.001/min at 340 nm.

### 1.3 Assay of adrenal $\Delta^5$ - $3\beta$ -HSD

Adrenal tissue was homogenized in a medium consisting of equal parts of 0.9% saline and 0.1 mol/L Na-phosphate buffer (pH 7.4) to give a tissue concentration of 4 mg/ml. Adrenal homogenate (1.6 ml) was incubated with 6 mg NAD in 0.2 ml phosphate buffer and 500 mg DHEA in 0.1 ml propylene glycol in a shaking incubator at 37°C for 90 min. After incubation, the mixture was immediately acidified with 0.1 ml of 3 mol/L acetate buffer (pH 5.0) and androstenedione as extracted with 10 ml ethyl acetate. This extract was dissolved in ethanol and the absorption was determined in a spectrophotometer at 240 nm according to the method of Rubin *et al.* (Rubin, 1961).

### 1.4 Assay of acid and alkaline phosphatase in sex organs

For quantitative estimation of alkaline phosphatase, decapsulated testicular tissue was homogenized in ice-cold homogenizing medium (0.22 mol/L Tris-HCl buffer pH 7.5) at a tissue concentration of 10 mg/ml. Prostatic tissue and seminal vesicular tissue were homogenized in the same medium at a tissue concentration of 20 mg/ml. 0.25 ml of the homogenate was added in a centrifuge tube containing 1 ml buffer (1 mmol/L p-nitrophenol phosphate in 1 mol/L tris buffer, pH 8). The mixture was incubated at 37°C for 30 min in a water bath. The assay was based on the formation of p-nitrophenol in the hydrolysis of p-nitrophenol phosphate. The activity was measured spectrophotometrically at 420 nm using Sicospec 100-A according to the procedure of Malamy and Horceker (Malamy, 1966).

For determination of acid phosphatase activity, the same homogenizing medium was used and the tissue concentration was as the same as above. Acid phosphatase activity was measured in acetate buffer of pH 5 using the p-nitrophenol phosphate as a substrate in presence of sodium fluoride and cobalt as specific activators (Venha-Pertulla, 1973).

## 2 Results

The body weight of the noise exposed group did not decreased significantly as compared with

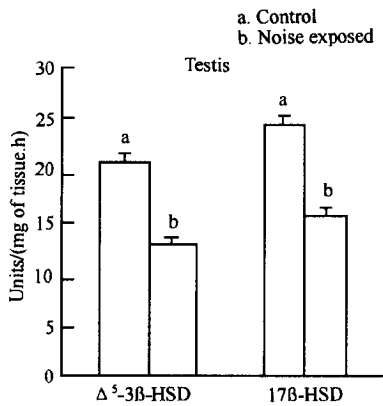
control (Table 1).

**Table 1** Effect of noise exposure (85dB) on body growth, gonadosomatic, prostatosomatic, seminal vesiculosomatic, adrenosomatic index and adrenal  $\Delta^5$ -3 $\beta$ -HSD activity in adult albino rats (values are mean  $\pm$  SE,  $n = 10$ )

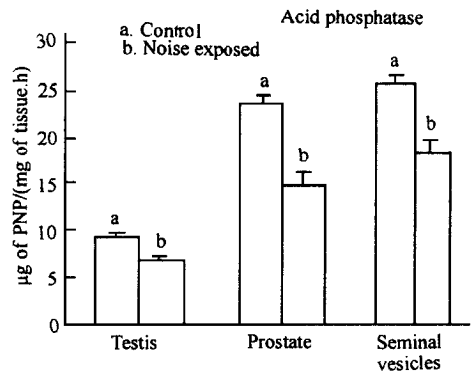
Group	Initial body wt, g	Final body wt., g	Gonadosomatic index, mg%	Prostatosomatic index, mg%	Seminal vesiculosomatic index, mg%	Adrenosomatic index, mg%	Adrenal $\Delta^5$ -3 $\beta$ -HSD activity, mol/mg of tissue/h
Control	120 $\pm$ 8 <sup>a</sup>	139 $\pm$ 9 <sup>a</sup>	1422 $\pm$ 20.62 <sup>a</sup>	215.14 $\pm$ 8.62 <sup>a</sup>	452.85 $\pm$ 18.32 <sup>a</sup>	1.23 <sup>a</sup>	20.52 $\pm$ 9.12 <sup>a</sup>
Noise exposed	124 $\pm$ 6 <sup>a</sup>	145 $\pm$ 8 <sup>a</sup>	1220 $\pm$ 33.33 <sup>b</sup>	155.62 $\pm$ 10.2562 <sup>b</sup>	380.32 $\pm$ 12.62 <sup>b</sup>	23.12 $\pm$ 1.92 <sup>b</sup>	29.32 $\pm$ 1.82 <sup>b</sup>

Notes: Figures with some superscript do not differ from each other significantly in each vertical column.  $P < 0.05$  (Student  $t$  test)

Testicular activities of  $\Delta^5$ -3 $\beta$  and HSD 17 $\beta$ -HSD were reduced significantly after noise exposure (Fig. 1). The acid phosphatase activities in testes, prostate and seminal vesicles were declined significantly in noise exposed group. The alkaline phosphatase activities in the tissues (Fig. 2) in contrast were elevated significantly in noise exposed group (Fig. 3). As shown in Table 1, the adrensomatic index and adrenal  $\Delta^5$ -3 $\beta$ -HSD activity were increased significantly in noise exposed group as compared with the control.



**Fig.1** Changes in testicular  $\Delta^5$ -3 $\beta$ -HSD and 17 $\beta$ -HSD activities after noise exposure (values are mean  $\pm$  SE of 10 animals in each group). Bars having different superscripts differ from each other significantly.  $P < 0.05$  (Student  $t$  test)



**Fig.2** Changes in acid phosphatase activities of testis and accessory sex organs after noise exposure (values are mean  $\pm$  SE of 10 animals in each group). Bars having different superscripts differ from each other significantly.  $P < 0.05$  (Student  $t$  test)

### 3 Discussion

In testicular steroidogenesis,  $\Delta^5$ -3 $\beta$  and 17 $\beta$ -HSD are key steroidogenic enzymes and their activities are regulated by gonadotrophins (Muroso, 1979). Noise exposure resulted a significant inhibition of the activities of these enzymes, that may be due to inhibition of pituitary gonadotrophins. Adrenal weight and adrenal  $\Delta^5$ -3 $\beta$ -HSD activity both were elevated significantly in the noise exposed group, which may be due to increase in ACTH, a stressful hormone secretion (Norman, 1997). The low plasma level of gonadotrophin in the noise exposed group may be due to high level of ACTH secretion as there is a reciprocal relationship between ACTH and gonadotrophin secretion (Selye, 1950; Nowell, 1957).

The output of sperm and the accessory fluids by both the testis and the accessory sex organs are greatly influenced by several enzyme activities (Mann, 1964).

Androgen controls the acid phosphatase activity in different reproductive organs (Levey, 1955; Tseng, 1986). Hypophysectomy and castration decreases the activity of those enzyme in sex

organs and testosterone supplementation results restoration(Ghosh, 1983; Tenniswood, 1978). So, diminution of acid phosphatase activity in testes, prostate and seminal vesicle in noise exposed group may be due to low androgen and this is supported by the inhibition in testicular  $\Delta^5$ -3 $\beta$  and 17 $\beta$ -HSD activity in noise exposed group. As accessory sex organ's weight is a good indicator for circulating level on androgen ( Moore, 1930 ), the decreased prostatosomatic index and seminal vesiculosomatic index in noise exposed group also support the possibility of the low plasma testosterone.

Alkaline phosphatase plays an important role in the formation of fructose from phosphorylate glucose (Pearse, 1969). This enzyme is widely distributed in different reproductive organs. The elevation in alkaline phosphatase activity in sex organs after noise exposure may be due to hyper adrenocortical activities (Cox, 1962; Saxena, 1980) and this is supported our result that noise exposure induces stimulation in adrenocortical steroidogenesis by the elevation of adrenal  $\Delta^5$ -3 $\beta$ -HSD activity.

Therefore, the above results provide evidence for the first time that noise exposure exerts an inhibitory effects on testicular steroidogenic enzymes and acid phosphatase activities in sex organs but stimulatory effect on alkaline phosphatase activities in sex organs and adrenocortical steroidogenesis. The underlying mechanisms involved and the eventual consequences of these deleterious effects of noise on reproductive organs remain to be elucidated in future experiments.

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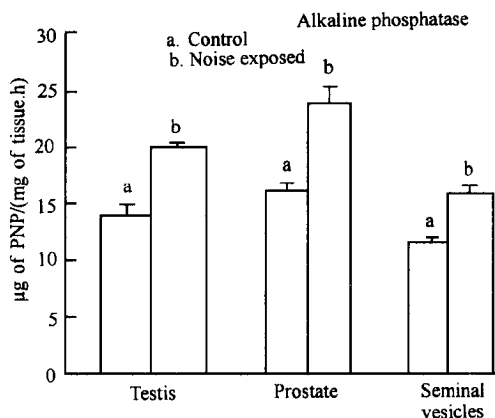


Fig.3 Changes in alkaline phosphatase activities of testis and accessory sex organs after noise exposure(values are mean  $\pm$  SE of 10 animals in each group). Bars having different superscripts differ from each other significantly.  $P < 0.05$  (Student *t* test)