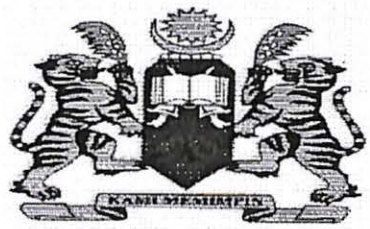
THE EFFECT OF NOISE STRESS ON THE SERUM TESTOSTERONE LEVEL IN SPRAQUE DWALEY RATS.



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KUBANG KERIAN,KELANTAN

2005

USM J/P- 06

BAHAGIAN PENYELIDIKAN & PEMBANGUNAN
CANSELORI
UNIVERSITI SAINS MALAYSIA

Laporan Akhir Projek Penyelidikan Jangka Pendek

1) Nama Penyelidik:
DR.G.CHANDRALEKHA.....
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Nama Penyelidik-Penyelidik Lain (Jika berkaitan) : ----
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2) Pusat Pengajian/Pusat/Unit : PPSP, DEPARTMENT OF ANATOMY.....

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3) Tajuk Projek: THE EFFECT OF NOISE STRESS ON THE SERUM TESTOSTERONE LEVEL IN SPRAQUE DAWLEY RATS.....

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4) (a) Penemuan Projek/Abstrak

(Perlu disediakan makluman di antara 100 – 200 perkataan di dalam Bahasa Malaysia dan Bahasa Inggeris. Ini kemudiannya akan dimuatkan ke dalam Laporan Tahunan Bahagian Penyelidikan & Pembangunan sebagai satu cara untuk menyampaikan dapatan projek tuan/puan kepada pihak Universiti).

Noise is a stress. Stress results in wide range of physiological responses in the body including the endocinological and reproductive functions. Only few studies evaluate the noise induced changes in the hormonal levels of rats and the results reported for correlation with the structural changes in the testis.

I have evaluated the impact of noise stress on the testosterone hormone .There were nine groups. Acute group was exposed to one hour and three hours noise of 100 dB of traffic noise. The chronic group has group a which has one hour exposure to 60days and one hour exposure to 90 days. The chronic group B has three hours of exposure to 60 days and 90 days. All have control. Each group has 6 rats.

After the stipulated time of exposure, the blood was collected for testosterone estimation and tissues like testis were collected. Testis was fixed in paraffin and sectioned and stained and observed for histopathological changes. Changes were noted in the testicular tubules and it is correlated with the decresed testosterone levels after exposure to noise stress. These changes differ from those reported earlier suggesting distinct adaptation potential in stressful condition.

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(b) Senaraikan Kata Kunci yang digunakan di dalam abstrak:

Bahasa Malaysia	Bahasa Inggeris
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5) Output Dan Faedah Projek

(a) Penerbitan (termasuk laporan/kertas seminar)
(Sila nyatakan jenis, tajuk, pengarang, tahun terbitan dan di mana telah diterbit/dibentangkan).r
presententing papers in conferences, publications

1. A PRELIMENARY STUDY OF THE EFFECT OF NOISE ON THE BEHAVIOURAL PATTERN OF THE MALE ALBINO RATS
1st Malaysian Conference On Aanatomical Sciences , 12-13 October 2002
USM',KELANTAN,**MALAYSIA(2002)**

2. 8TH NATIONAL CONFERENCE ON MEDICAL SCIENCES ,8-9,May 2003 , UNIVERSITI SAINS MALAYSIA, KELANTAN, **MALAYSIA** "changes observed in the histological pattern of the adrenal gland after exposure to noise in albino rats".

3. 9TH NATIONAL CONFERENCE ON MEDICAL SCIENCES , 22-23, May 2004 , UNIVERSITI SAINS MALAYSIA, KELANTAN, **MALAYSIA**
Chandralekha,Jeganathan ,Evaluation of Noise stress effect on serum corticosterone and leptin levels in Albino Rats.

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(b) Faedah-Faedah Lain Seperti Perkembangan Produk, Prospek Komersialisasi Dan Pendaftaran Paten.

(b) Faedah-Faedah Lain Seperti Perkembangan Produk, Prospek Komersialisasi Dan Pendaftaran Paten.
(Jika ada dan jika perlu, sila guna kertas berasingan)

Publication in the journals, teaching for M.D-students and postgraduate, M.Sc in Clinical anatomy.

It I s used for research training and project works for undergraduate and postgraduates.

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(c) Latihan Gunatenaga Manusia

i) Pelajar Siswazah: The results of the research were presented to the MD students at the lectures on endocrine,reproduction, environmental changes in relation to above systems.....

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ii) Pelajar Prasiswazah: ...The results of the research were presented to MSc clinical antomy student, and postgraduate students in USM, and by telemedicine to postgraduate students in Penang, Malaca students

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iii) Lain-Lain : .....

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6. Peralatan Yang Telah Dibeli:

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UNTUK KEGUNAAN JAWATANKUASA PENYELIDIKAN UNIVERSITI

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T/TANGAN PENGERUSI
J/K PENYELIDIKAN
PUSAT PENGAJIAN

Professor Zabidi Azhar Mohd. Hussin
Chairman of Research & Ethics Committee
School of Medical Sciences
Health Campus
Universiti Sains Malaysia
16150 Kubang Kerian,
KELANTAN, MALAYSIA.

Abstract No. O-25

Title : A PRELIMENARY STUDY OF THE NOISE STRESS EFFECT ON THE BEHAVIOURAL PATTERN OF THE MALE ALBINO RATS

Authors : G. Chandralekha and R. Jeganathan*

Institution : Department of Anatomy, School of Medical Sciences, Universiti Sains Malaysia; *Department of Anatomy, Stanley Medical College, Chennai-I, India.

Introduction : The noise, one of the environmental pollutants, is considered as a strong stress factor. The environmentalists consider it as a type of pollution similar to air pollution, water pollution etc. Works were done on cardiovascular, respiratory, endocrine, and reproductive systems and on behavior. This preliminary study was designed to see the behavioral changes of the albino rats when they are exposed to noise.

Objectives : To assess the effect of noise exposure (generator noise-100 decibel (dB) on the behavioral pattern of the male albino rats.

Methodology : The animals were grouped as control , acute and chronic. Acute group was exposed to one hour, and three hours of generator noise exposure of 100dB continuously/day. The chronic groups were exposed to one hour, and three hours of continuous noise exposure/day for 30 days and 45 days. The animals were keenly observed for their reaction towards the noise stress.

Result : Annoyance, disturbance, loud cry were the common features noticed when the rats were exposed to acute noise stress of 100 dB. The rats remain confined in the cage and refused to take food and water in chronic groups. When the animals were exposed to the Grip Strength test, there was significant decline in the grip of the rats belong to the acute and chronic groups. In the Splay test, the distance between fore limbs was significantly increased after exposure to noise stress of 100dB, in acute and chronic groups. The increase was time bound. 100 dB of noise stress has significant effect on the splay and grip strength of the rats, in acute , as well as in chronic groups.

Discussion & Conclusion : Noise has definite effect on the behavioural pattern of albino rats.

ABSTRACTS

Results and Conclusions :

1. *The chromatolysed neuron somata (representing the motor neuron somata of median nerve) were found to be located in the caudal-third to caudal-half of C-6, the whole lengths of C-7, C-8 and T-1 and the cranial part (0.8 to 2.5 mm)of T-2.*

2. *In C-6 and C-7, these chromatolysed neuron somata were located in the dorsal part of dorsolateral column (DL) and in C-8 and T-1 they were located in all parts of retrodorsolateral (RDL) and DL of the ventral grey horn. In T-2, they were located in RDL only.*

ABSTRACT CODE (O - C3 - 3)

Title : CHANGES OBSERVED IN THE HISTOLOGICAL PATTERN OF THE ADRENAL GLAND AFTER EXPOSURE TO NOISE IN ALBINO RATS".

Authors : Dr.G.Chandralekha & Dr.R.Jeganathan*

*Institution : Leaturer in Anatomi, USM, Kelantan, Malaysia ; * Ass.Professor of Anatomi Stanley Medical College,Chennai,India.*

Abstract : Human tissues are vulnerable to stress factors. Like heat stress, cold stress ,swimming stress ,noise is also considered to be a stress factor. 'Noise is a silent killer' stated by Ogale(1999). Pituitary adrenal activity in rats were affected by 85 dB chronic noise stress (Armario and Castellanos ,1984). The structural pattern of the adrenal gland when exposed to noise was not focused much. So in the present study the animals were exposed to 120 db of generator noise and its effect on the structural pattern was observed. The animals were grouped as acute and chronic. The acute group was exposed to 1hour, 2 hours, and three hours noise exposure(120dB) and the chronic groups were exposed to 30 days, 45 days and 60 days of daily one hour noise (120dB)exposure.The animals were given barbiturate injection and sacrificed and adrenal glands were collected and processed for paraffin section. The adrenal tissue shows overall swelling, vascular congestion, lipid depletion in acute and chronic noise exposure in varying degree.

ABSTRACT CODE (O - C3 - 4)

Title : INITIAL INTERACTION OF RAT MESENCHYMAL CELLS WITH A CORAL DISC

Authors : K. A. AL-SALIHI, A. R. SAMSUDINE, SHAMSURIA OMAR

Institution : SCHOOL OF DENTAL SCIENCES, UNIVERSITI SAINAS MALAYSIA

OBJECTIVE

This study was designed to investigate the surface attachment and morphology of rat bone marrow stromal cell on the surface of coral shaped as disc using laser scanning confocal microscopy.

Introduction : The surface of implant materials presented to cells can be considered as a foreign chemical species with reactive sites. Substrate surface characteristics have been shown to affect many aspects of cellular behavior, such as attachment, spreading, proliferation and differentiation. Cells interact with a biomaterial through a layer of extracellular matrix (ECM) proteins. This layer may be adsorbed from fluids surrounding the material, or may be synthesized by the cells. These proteins act as ligand for a family of cell surface proteins called integrins. These receptors consist of two transmembrane subunits, i.e. alpha and beta subunit. The intracellular domain of the beta subunit associates with cytoskeletal and

POSTER 95

EVALUATION OF NOISE STRESS EFFECT ON SERUM CORTICOSTERONE AND SERUM LEPTIN LEVELS IN ALBINO RATS.

Authors : *G.Chandralekha* and R. Jeganathan***

Institution : **Department Anatomi, Univerustu Sains Malaysia, Kelantan*
*** Associate Professor of Anatomi, Stanley Medical College, Chennai-1, India.*

Introduction :

Noise stress produces significant physiological and biochemical changes in animals as well as in humans The damaging effects of noise on hearing have been extensively studied. However very little information is available regarding the effect of noise on other body functions . Similar to other types of stress, noise stress has also been shown to increase the levels of stress hormones like cortisol and nor epinephrine.

Objectives :

The objective of the present study is to evaluate the effect of acute and chronic noise stress on albino rats and to evaluate the serum corticosterone and serum leptin levels.

Methodology :

The present study examined the effect of exposure of albino rats to acute and chronic noise stress on two important interlaced endocrine levels of serum leptin and corticosterone. In acute experiments ,the animals were exposed to 120 dB noise for the duration of 1, 2 or 3 hrs . In chronic experiments, the animals were exposed to 120dB of noise for one hour per day for 30, 45 and 60 days. Serum corticosterone and leptin levels were measured in these animals. The results were analyzed using Anova followed by Dunne's test. The 'p' <0.05 was considered statistically significant.

Results :

The results indicated significant elevation in the levels of serum corticosterone and serum leptin after exposure to noise stress in the above mentioned paradigm.The acute noise stressed animals for one hour, two hours and three hours showed significant elevation in serum corticosteriod and in serum leptin levels. The chronic exposed animals one hour exposure to noise stress per day to 30 days, 45 days and 6odays, also showed significant elevation in serum corticosterone and in serum leptin levels.. Exposure to stressors like noise can be expected to induce alterations in the level of leptin.

Conclusion :

In conclusion the present study clearly indicate that sustained exposure to noise stress results in significant elevation in serum corticosterone and in serum leptin levels. These two hormones have wide ranging effects on metabolism, growth and reproduction . Hence, continuous exposure to noise stress may have many adverse effects on these vital physiological functions for which the alteration in the level of these two hormones may play a significant contributory role. However, this present study may be the first of its kind to report the elevation in leptin levels after exposure to noise stress. The elevation in the levels of corticosterone and leptin after exposure to noise stress may alter the vital process in metabolism, growth and reproduction, which are the main functions of these two hormones.

ACKNOWLEDGEMENT

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I acknowledge The Research Creativity and Management Office, Chancellory, Universiti Sains Malaysia, 11800, Penang, Malaysia.

2. R&D department , Universiti Sains Malaysia, 16150, Kubang kerian, Kelantan,

3. The Chairman,Animal Ethics, School of Medical Scienceal Committee , USM, Kubang Kerian, Kelantan

4. I thank Dean, Deputy Deans and Deputy Dean of Research for their kind help and cooperation.

5. I thank Head of the department of Anatomy for his kind support.

6. I thank Staff of R&D department , Anatomy department, Physiology Department, Pathology department, Immunology departments of PPSP and the Research department of PPSG, Kubang kerian, Kelantan for their help and guideness and moral support.

7. My special thanks to Mr.Harissal, Technologist, department of Anatomy for his kind cooperation, effort and help to finish this project.

8. I thank my husband Dr.Jeganathan and son JC.Charan for their help, moral support, and valuable suggestions to finish this project .

NOISE STRESS EFFECT ON SERUM TESTOSTERONE AND ON TESTICULAR MORPHOLOGY

Chandralekha G*, Jeganathan R**, Charan JC***

*Anatomy department, Universiti Sains Malaysia,Kelantan.**Vice Principal And Head of the department of Anatomy, Ragas Dental College, Chennai, India, ***Dr.MGR Medical and Research College (Deemed university) Chennai,India.

INTRODUCTION AND REVIEW

Collins 1995, indicated that noise is also partially responsible for reduced reproductivity Armario, et al 1887 , stated that the pituitary gonadal response to stress would be a more sensitive index of abnormalities induced by protein calorie deficit than basal concentration of LH, follicular stimulating hormone (FSH) or testosterone. Armario and Castellonas 1984, reported that acute noise stress of 80 dB, increased the testosterone level. Gopalakrishnan et al 1998, indicated that tamoxifen decreased the serum testosterone level and disrupted the testicular seminiferous epithelium. Tamoxefen citrate induces multinucleated giant cells and germinal epithelial sloughing in a dose dependent manner (Urban J.A.D'Souza 2003) On morphometric analysis of testicular tissue it was observed that there was no significant change in spermatogonia and spermatocytes, but significant decrease in spermatids and sperm number as well as daily

sperm production in the immunized male rats. The epididymal spermatozoa were markedly reduced in number and were largely found to be agglutinated. Elevated serum FSH is also considered to be a reliable indicator of germinal epithelial damage and is usually associated with severe oligospermia or azoospermia. *(Matsumato and Bremner, 1989).*

AIM

Very few reports on the noise effect on seminiferous morphological changes and its correlation with testosterone were available .Hence the aim of the present study was to examine the effects of noise stress on the seminiferous tubules and testicular morphology and on the serum testosterone levels and their correlations.

MATERIALS AND METHODS

Male Spraque Dewley rats obtained from Animal house unit of Universiti Sains Malaysia, Kelantan were used in this study. All animals were of the same age group and weighing 100 – 120 g. The rats were housed in standard polyprophylene cages with laboratory chew and water *ad libitum*. The animals were kept in under normal room temperature. The care and maintenance of animals were as per the approved guidelines. The animals were randomly divided into acute and chronic groups. In acute group, the

animals were exposed to one hour and three hours exposure to noise stress. And in chronic group, the rats were exposed to continuous noise stress for one hour and three hours for 60 days and 90 days. There were 9 groups. Each group consisted of six animals. Both acute and chronic groups were exposed to 100 dB of recorded traffic noise for the periods mentioned above. Upon completion of all noise exposure regimens, the animals were anesthetized with an intraperitoneal injection of intraval sodium (60 mg/kg body weight) and exsanginated after collection of blood for hormonal assay.

On the stipulated day, after the collection of the blood for hormonal assay, laporatomy was performed; the testis and epididymides were carefully removed and separated. Tunica vaginalis was carefully removed and the testes were dissected out and cleaned with cold physiological saline to remove blood and the adhering tissues. The samples were then fixed in 10% formol-saline and in fresh alcoholic Bouins fluid for 8 hours, and embedded in paraffin. 5μm sections were cut and stained with haematoxyllin and eosin. The testis histopathology was performed according to Sobarzo and Bustos-Obregon and Russel et al. The tissue sections were observed under a light microscope for qualitative study.

RESULT

There was a significant reduction in serum testosterone level in one hour and three hours acute noise exposed rats. Similar reduction was also noticed in one hour and three

hours of 60 days and 90 days noise exposure.in chronic group. There was a gradual

reduction in testosterone level and maximum reduction was seen after three hours of 90 days exposure.(Table1, Chart 1). But there was an increase in 60 days of 3 hours noise exposure when compared to three hours in acute group.

Histopathological changes in testis and epididymis were observed in animals subjected to both acute and chronic noise stress. The severity of changes was also dependent on the duration of exposure to noise stress. The section taken from testis of rats subjected to one hour, and three hours in acute showed normal histological architecture. After 60 days of chronic noise exposure in one hour and three hours duration the normal arrangements were maintained in the periphery. But there were changes in the tubules. Some of the tubules were separated from each other.

Maturation arrests in spermatogenesis were observed in some of the tubules. Even after 90 days of exposure to noise for one hour and three hours, the same picture was maintained as in 60 days. But some tubules showed interruption in the basement membrane. (Picture 2) The basement membrane was thinned out in some tubules. Degenerative changes were noted in some tubules. There were maturation arrests seen in few seminiferous tubules.

There were changes in the basement membrane which was disturbed in some places. The

contents of the lumen were sloughed out side the tubes. (Pictue2).

STATISTICAL ANALYSIS

The data of the results was subjected to ANOVA followed by Dunnett's "t" test for statistical significance. A p level of < 0.05 was considered statistically significant

Discussions

There was a significant reduction in serum testosterone level in acute noise and chronic noise exposed group. There was a gradual reduction in testosterone level and maximum reduction was seen after three hours of 90 days continuous exposure to noise. Lutenizing hormone normally acts on the testes and increases the plasma testosterone level. Surprisingly in the present study, there was a significant reduction in serum testosterone levels. These effects were noticeable in both acute and chronically noise stressed animals. The above observation in the present study though appears paradoxical, is not uncommon. Similar observations have been recorded earlier in mutant rats by *Noguchi et al (1993)*. They reported that in certain strain of mutant rats, the plasma concentration of follicular stimulating hormone (FSH) and lutenizing hormone (LH) were significantly higher than in normal rats and surprisingly, plasma

concentration of testosterone was lowered in these animals. These results suggest that the feed back action of testosterone on the hypothalamus and pituitary gland may be impaired. However, it is well established that testicular inhibin is one of the major factors controlling the secretion of FSH in male rats. *(De Jong 1988).* The hyper secretion of FSH in the mutant rat may be related to insufficient secretion of inhibin by sertoli cells. In addition, the hyper secretion of LH was also demonstrated in the mutant rats from 10 weeks of age. These results also suggest that the secretory response of leydig cells of the mutant rats were slightly decreased since plasma concentration of testosterone did not increase in *response to high concentration of plasma LH.*

Noguchi et al (1993) also reported many abnormalities in the testicular histology of mutant rats. Tubules containing only spermatogonia , and in some sertoli cells were prominent in the seminiferous epithelium of mutant rats. Similarly, in the present study, the histology of testes in rats exposed to 100dB of noise stress indicated few spermatogonia in seminiferous tubules. Maturation arrest was a prominent feature in the germinal epithelium in the present study. The basement membranes were interrupted. These changes were appearing gradually and the general plan of arrangement of the architecture was altered in the center rather than at the periphery. These observations on hormonal changes and testicular histology raises the question, '*whether noise stress can*

lead to any mutational changes in male reproductive system?' This aspect has to be viewed seriously since the younger generation is exposed to noise stress overwhelmingly. Elevated serum FSH is also considered to be a reliable indicator of germinal epithelial damage and is usually associated with severe oligospermia or azoospermia. *(Matsumato and Bremner, 1989).* After 60 days of chronic noise exposure, maturation arrest of spermatogenesis was noticed in rats. This effect became very prominent after 90 days. Eventhough, serum FSH level was elevated , oligospermia or azoospermia were not observed by the authors. The sperm concentration and percentage of live sperms were decreasing after noise stress. There is a clear evidence that the motility of the sperm is severely affected in noise stress exposed ats(Chandralekha,2002). It has been reported that young men may experience alterations in sperm quality (fewer motile sperms, abnormal sperms) after exposure to periods of elevated air pollution, by *Selevan et al, 2000,* which can also be considered a form of stress.

The main function of testosterone is in the regulation of the development of the spermatozoa, the growth, development and maintenance besides its action on accessory reproductive glands and maintaining the secondary sexual characters. The testis has two interrelated functions, the production of sperms (spermatogenesis) and production of steroidogenesis like secreting the testosterone hormone. When testosterone levels were

decreased, the maturation arrest were noted in the seminiferous tubules. Both the effects are due to the noise exposure. Hence, it may be possible that exposure to noise over long periods of time, can also result in maturation arrest because of decreased testosterone levels as observed by the author in the present study and also reported by *Selevan et al, (2000)*.

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THE EFFECT OF NOISE STRESS ON THE SERUM TESTOSTERONE LEVEL IN SPRAQUE DAWLEY RATS.

1. INTRODUCTION:

Noise is a known stressor. According to Lai 1987 , short term exposure to noise is unavoidable but long term noise exposure is detrimental to health. *Collins 1995, indicated that n*oise is also partially responsible for reduced reproductivity .Cosa and Cosa s 1989, tated that noise stress was related with neuroendocrinological response. Moreover the pituitary adrenal activity in rats were affected by 85 dB of chronic noise stress as stated by Armario and Castellanos jm 1984. Both lutenisizing hormone(LH) and testosterone responses to acute noise stress were impaired by water restriction. Armario, et al 1887 stated that the pituitary gonadal response to stress would be a more sensitive index of abnormalities induced by protein calorie deficit than basal concentration of LH, follicular stimulating hormone (FSH) or testosterone. The acute noise stress showed increase in testosterone level in goats

(Kobegenova, et al, 1985) and rats (Prabhakaran et al, 1988 and De Boer, et al, 1988).Jean Saldaha 1999, reported that noise stress inhibits the production of seratonin, which affects the sperm count and causes piddling ejaculation in human . Armario and Castellonas,1984 stated that acute noise stress of 80 dB, increased the testosterone level . Edmouders, et al, 1979 in their report stated that congenital effects was observed in the offspring of the people working at the industries . Fletcher and Busnel, 1978, reported that mice exposed to 105 dB of intermittent noise for 4 hours per day exhibited longer time interval between litters and low weight gain of young compared to control. Noguchi et al (1993) also reported many abnormalities in the testicular histology of mutant rats. Gopalakrishnan et al , indicated that tamoxifen decreased the serum testosterone level and disrupted the testicular seminiferous epithelium . Urban J A D'Soza 2003, also reported that the tamoxefen citrate induces multinucleated giant cells and germinal epithelial sloughing in a dose dependent manner. Mice exposed to 105 dB of intermittent noise for 4 hours per day exhibited longer time interval between litters and low weight gain of young compared to control (Fletcher and Busnel, 1978). On morphometric analysis of testicular tissue it was observed that there was

no significant change in spermatogonia and spermatocytes but significant decrease in spermatids and sperm number as well as daily sperm production in the immunized male rats. The epididymal spermatozoa were markedly reduced in number and were largely found to be agglutinated. There were only few reports depicting the increase or decrease of testosterone level in relation to exposure to noise at various decibels. Very few reports were available on the testosterone effect on the testicular morphology. Hence the aim of the present study was to examine the effects of noise stress on the testosterone levels and on the seminiferous epithelium and testicular morphology.

CHAPTER ONE

1. INTRODUCTION AND LITERATURE REVIEW

The civilizations of the technologically advanced present society has a new kind of stress exposure namely ***'noise'***. Of all types of environmental pollutants, noise is the most prevalent and insidious natural pollutant with deleterious physiological and psychological effects. The subtle and insidious nature of noise makes it a **'slow agent of death'.** Knowingly or unknowingly we are exposed to different kinds of noise in occupations, environments and in communities. Now health scientists are realizing the adverse effects of exposure to noise on human health. A sudden noise striking the ear produces myriad of physiological change. Noise when considered as a stressor, can affect various physiological system in the body.

In ICMR bulletin 1996, it is stated that noise is a growing problem in

developing countries. Samuel Rosen 1991 , had reported that noise affects all body systems whether we like it or not . An American ENT surgeon said, "You may forgive the noise, but your body never will ". Lang man 1996 , stated that even the fetus in the mother's womb is vulnerable to excessive noise leading to congenital defects in fetal nervous system and that may affect the behavioral pattern in later life. *So Ogal, 1999, stated that "Noise is a silent killer"*

1.1 Noise as stress

In fact, noise is considered as a strong stress factor for human beings and animals. The environmentalists consider it, as a type of pollution similar to air pollution, water pollution etc. Similar to other types of stressors, noise is also reported to produce many changes in the biological system. Hence, it is considered as a stressor. Even though the primary target of noise on the ear, other body system are also adversely affected. As stated by Lai,1987, short term noise is unavoidable but long term noise exposure is detrimental to health.

1.2 Properties of sound

There are two important properties of sound, the *pitch* or frequency and the *intensity* or loudness. The pitch is the rate of vibration of the sound and is measured in Hertz (HZ) unit. The intensity or loudness of the noise is based on the physical unit and is measured in decibels .

1.3 Unit of measurement

Noise is measured by a unit called decibel which is named after Alexander Graham bell. The decibel is a physical unit based on the weakest sound that can be detected by the human ear. A decibel (dB) is one-tenth of a bel. *(Manorama, 1996).* The decibel scale is logarithmic, i.e. for every increase of 1 dB, it increases by 10 times. The human ear cannot hear frequencies higher than 20,000 vibrations/second or 20,000 Hz. This inaudible sound is called ULTRASONIC sound and bats produce such ultrasonic sounds. The speed of sound in air at sea level is 331 meters/second. In water it is 15 times faster than in air. The supersonic speed is 1216 km/hour and it is measured in ' MACH ' named after German physicist Ernest Mach.

1.4 Types of noise

Noise is produced by vibration of an object or mechanism which is transmitted in the form of waves, with alternating increase and decrease in pressure . There are two types of noise.

1.A continuous shrill noise (pure tone noise 92 -98 dB

e.g., a siren 92 -98 dB)

2. White noise (or an intermittent heavy artillery noise static on

the radio 102 dB, Shanker *et al 1999).*

Dixit 1991, stated that human ear can safely respond to pressures up to 85-90 dB . Any intensity higher than that is harmful and can cause hearing impairment. The maximum exposure to out door noise is 120-140 dB, and Very rarely to 170 dB. The maximum exposure to indoor noise is 60 –80 dB. So , human being is exposed to average noise of 100 –110 dB daily. Our feasible auditory capacity is 85 – 90 dB, but daily we are exposed to 15-20 extra decibels. In course of our life time, such an exposure can result in serious damages to our body functions.

1.5 Source of Noise:

Noise sources are many. But the present study is on the road traffic noise. The back ground environmental noise levels rise is in direct proportion to the population of a given area. The noise appears to be endemic in urban condition. In the cities, the main causes are due to traffic noise such as motors, and exhaust system of automobiles, trucks, buses, and two wheelers. This type of noise may be exacerbated in the narrow streets where sound reverberates between the buildings.

1.6 Adverse effects of noise stress

There were many adverse effects. The annoyance, loud cry, disturbances and damage caused by noise is classified into two.

a. The effect on auditory apparatus.

b. The effect on other parts of body.

1.6 a. Effects on auditory apparatus.

Extensive work had been conducted on the effects of noise on the auditory system like acoustic trauma, auditory weariness, noise deafness and the vestibular system like vertigo, sickness, trouble of balance. Those lesions happened after prolonged exposure to 80 – 85

dBand the cilia of the hair cells of the internal ear becomes flattened and the death of cilia along with loss of hearing ability of the ear (Ogale, 1999). But exposure to air craft noise causes vertigo and hearing impairment . But Ames and Arehart, 1972 reported that exposure to 140 dB, 1400 HZ increases audiogenic seizures in men The deafness, rupture of tympanic membrane, death of hair cells, dislocation of ossicles, acoustic trauma were also reported after exposed to 150 dB.

1.6 b Effects on other parts of body

The non-auditory effect of noise stress has taken important move in recent years. Most of these studies documented revealed the alterations in the levels of cortisol, adrenocorticotrophin (ACTH) and nor-epinephrine, which are primarily considered as stress hormones. Any type of stress can also alter the behavioral responses and this has been confirmed by some of the earlier studies.Irwin et al 1989, reported that the analysis of the association between physiological and behavioral measures revealed that the degree of noise induced suppression of both general activity and ingestive behaviors significantly correlated with activation of adrenal steroid secretion following both acute and the repeated noise exposures

It was also reported in the literature that exposure to

aircraft noise of 100 – 120 dB per 12 hr daily on sows, boars, were not affected neither physiologically nor in their behavior. (Douglas, et al 1963). Metabolism is the fundamental process of energy production and maintenance of life. It is regulated by the hormones of endocrine glands. When the rats were maintained on normal magnesium containing diet for 12 weeks, subjected to audiogenic sound of 85 dB 12 hours per day for 8 weeks, 95 dB 16 hours per day for 4 weeks, showed a significant elevation in systolic and diastolic arterial blood pressure (ABP). Altura et al 1992 stated that the plasma magnesium showed a 15% deficit and elevation in calcium .

The noise stress effect was also studied on eyes which are the organ for sight. The rods and cones are responsible for day and night vision. The threshold performance of dark adapted cone and rod vision can be consistently described as noise limited. (Donner, 1992).

1.6 .b.1 Noise effect on endocrine and reproduction

Collins,1995, stated that noise also partially responsible for increase of consumption of alcohol, social handicap, reduced reproductivity, decreased performance in learning, absenteeism in school and work place increased drug use, accident, violent behavior, and manifesting like

anxiety behavior. Cosa and Cosa, 1989 also reported that these are related with neuroendocrinological response to the noise impact.

Both lutenisizing hormone(LH) and testosterone responses to acute noise stress were impaired by water restriction. Armario and Castellanos, 1984. stated that pituitary adrenal activity in rats were affected by 85 dB . Armario, et al 1887 also ated that the pituitary gonadal response to stress would be a more sensitive index of abnormalities induced by protein calorie deficit than basal concentration of LH, follicular stimulating hormone (FSH) or testosterone chronic noise stress . But there was reduction in ACTH in noise exposed rats to chronic stress .Armario and Castellanos1984; De Boer ,et al 1989, also stated that after repeated exposure to noise stress, the plasma corticosterone, adrenaline and nor-adrenaline were reduced but increased in acute noise stress exposure (De Boer, et al, 1988). But Kobegenova et al 1985, had reported that the acute noise stress in goats showed increase in testosterone level.and rats (Prabhakaran et al, 1988 and De Boer, et al, 1988).

Significantly decreased pregnancy rate and embryo lethal effect

was observed in mice exposed to high frequency of noise (Popper and Tarolga,1981). Jean Saldaha,1999, stated that occupational noise stress is geniotoxic agent. Noise stress inhibits the production of seratonin, which affects the sperm count and causes piddling ejaculation in human .

Nagalakshmi et al 1999, reported abnormalities in reproduction due to occupational noise stress . Acute noise stress of 80 dB, increased the testosterone level (Armario and Castellonas,1984). Congenital effects was observed in the offspring of the people working at the industries (Edmouders, et al, 1979). Mice exposed to 105 dB of intermittent noise for 4 hours per day exhibited longer time interval between litters and low weight gain of young compared to control (Fletcher and Busnel, 1978).

There were literature available about the noise effect on cardiovascular system, respiratory system, body metabolism, behavior, biochemical aspects, on rods and cones, on endocrines and on reproductive system. But very literatures were available about the correlation between the hormonal effect on the testicular morphology. Hence the present study was undertaken.

CHAPTER 2

2. OBJECTIVES

The aim of the present study is to investigate the acute and chronic exposure to 100db of noise on

i. the serum testosterone level in Spraque Dawley rats .
ii. on the histopathological changes in testis .
iii. and to correlate the hormonal changes with structural changes in the testicular morphology.

CHAPTER 3

3.MATERIALS AND METHODS

3. 1. Animals:

Male Spraque Dawley rats obtained from Animal house unit of Universiti Sains Malaysia ,Kelantan were used in this study. All animals were of the same age group and weighing 100 – 120 g. The rats were housed in standard polyprophylene cages with laboratory chew and water ***ad libitum***. The animals were kept in under normal room temperature. The care and maintenance of animals were as per the approved guidelines .

3.2 Inclusion criteria

i. Male rats

ii. Weight 100-120gm

iii . 100 dB of recorded traffic noise

3.3 Exclusion criteria

I .Female rats

ii. Weight below 100gm

iii. Weight above 120 gm

iv. Above 100dB or below 100 dB of recorded noise .

v. Unrecorded noise .

3.4 Sample size

The sample size for the present study were calculated based on the study done by Kobegenova, et al, (1985), Prabhakaran et al, (1988) and De Boer, et al, (1988), Archana and Namasivayam (1999). The sample size was calculated with the confidence interval was 95% and precision was 5% and it was approved by the animal ethics committee.

3.5 Animal ethics approval

This study was approved by The Universiti Sains Malaysia Health Campus Animal Ethics Committee, Kelantan. Ref: PPSG/07(A)/044 ,2 Disember 2002 [copy].

3.6 Pilot study

As stated by Dickens 1973 , the rats which were responding to normal sound was selected. The animals which were placed in a cage was tested by considering the ear or head or body movement towards the source of sound for the pilot study. The animals used for chronic experiment were periodically tested every week during the course of experimentation.

3.7 Groups

The animals were randomly divided into acute and chronic groups . There were 9 groups. Each group consisted of six animals .

I. Acute groups

The acute groups were subdivided into ,

a.Control

b.One hour exposure to noise.

c. Three hours exposure to noise continuously.

ii. Chronic groups

The chronic groups were also subdivided into A and B .

A. group- one hour noise exposure daily

i.. control

ii. one hour noise exposure daily for 60 days

iii. one hour noise exposure daily for 90 days

B Group-.three (3) hours noise exposure daily

i.control

ii.for 60 days

iii. for 90 days.

3.8. Method of exposure to noise:

The animals were kept in a sound proof room during experiment. Both acute and chronic groups were exposed to 100 dB of recorded traffic noise for the periods mentioned above. Their normal habitats were maintained. The animals were exposed to a pure tone noise of sine waves with a frequency of 10,000 Hz and an intensity of 100 dB. A sound level decibel meter was used to measure and maintain the intensity of noise. The controls were maintained and treated like experimental groups in all aspects except exposure to noise .

3.9 Method of sacrifice:

Upon completion of all noise exposure regimens, the animals were anesthetized with an intraperitoneal injection of intraval sodium (60 mg/kg body weight) and exsanginated after collection of blood for hormonal assay

3.10 Blood-for hormonal assay

Before the animals were exanginated, approximately 5 ml of blood was collected from each rat for hormonal assay.

3.11 Hormonal assay

Testosterone hormone was assessed by using Elisa method. The DRG Testosterone ELISA KIT was used which was based on the competition principle and the microplate separation. The separated serum were stored at 2-8° C for up to 24 hours. The reagents and the specimens were brought to the room temperature before use. After the assay was completed ,the results were recorded immediately.

3.12 Tissue preparation and Histopathology

On the stipulated day, after the collection of the blood for hormonal assay, laparatomy was performed and both the testis were carefully removed . Tunica vaginalis was carefully removed and the testes were dissected out and cleaned with cold physiological saline to remove blood and the adhering tissues. The samples were then fixed in 10% formol-saline and embedded in paraffin. Few tissues were fixed in fresh alcoholic Bouin's fluid for 8 hours, embedded in paraffin and 5μm sections were cut and stained with haematoxyllin and eosin. The testicula histopathology was performed according to Sobarzo and Bustos-Obregon . The sections were observed under light microscope.

CHAPTER 4

4.RESULTS

4.1 Hormonal assay –Testosterone

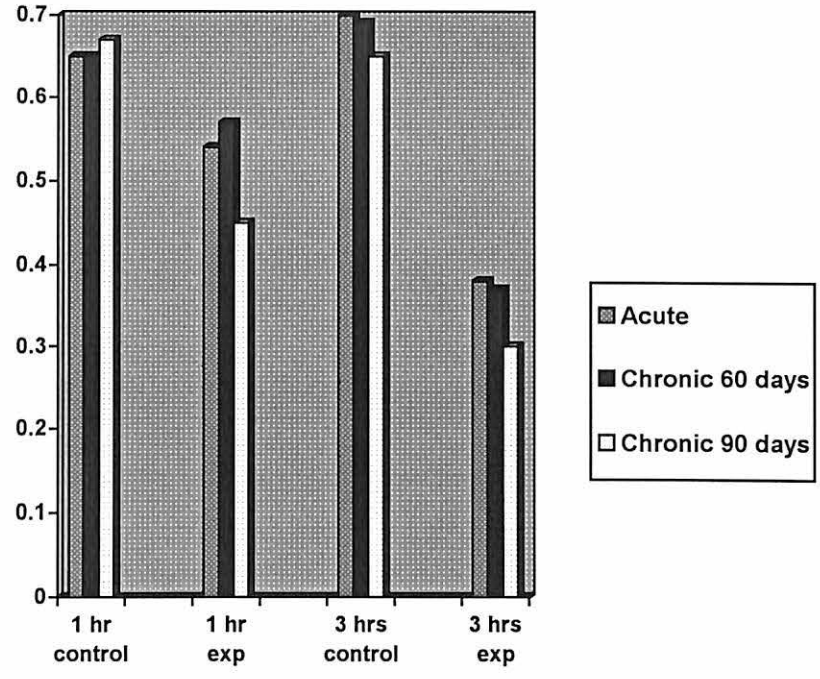
There was a significant reduction in serum testosterone level in one hour and three hours acute noise exposed rats. Similar reduction was also noticed in one hour and three hours of 60 days and 90 days noise exposure in chronic group. There was a gradual reduction in testosterone level and maximum reduction was seen after three hours of 90 days exposure (Table1, Chart 1).

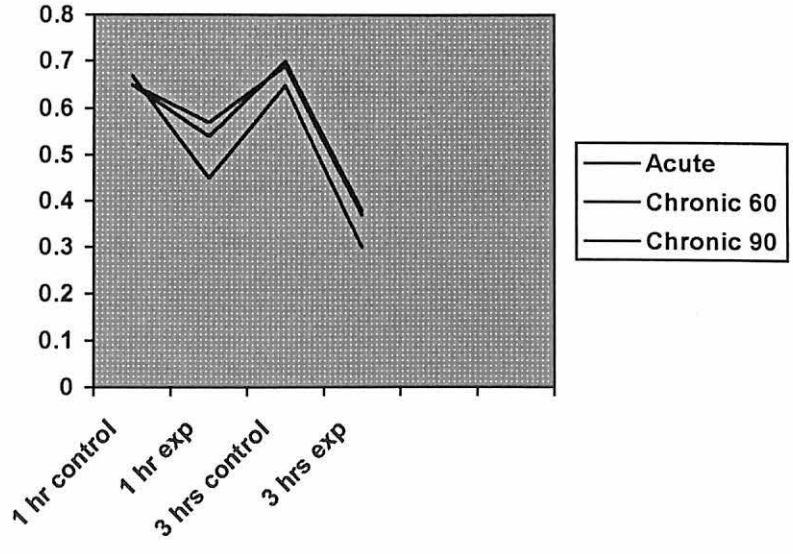
Table 1

The Effect of noise stress on sr. testosterone level in Spraque Dawley rats.

Period of exposure to noise	**Testosterone (ng/dl) Control**	**Testosterone (ng/dl) Experiment**
Acute noise exposure		
I hour	0.65±0.01	0.54±0.002**
3 hours	0.70±0.02	0.38±0.025**
Chronic noise exposure		
60days-I hour	0.65±0.01	0.37±0.016**
90days-1 hour	0.67±0.03	0.45±0.012**
60days-3hours	0.69±0.04	0.57±0.020**
90days-3 hours	0.65±0.04	0.30±0.01**

**$p<0.01$ when compare to respective control value





SERUM TESTOSTERONE LEVEL AFTER EXPOSRE TO NOISE STRESS.

4.2 Histopathological Changes

Marked histopathological changes in testis was observed in animals subjected to both acute and chronic noise stress. The severity of changes was also dependent on the duration of exposure to noise stress.

4.3 Histopathological observations in testis after acute noise stress

The section taken from testis of rats subjected to one hour noise stress, showed normal histological architecture as observed in control animals. The testis had many lobes and each lobe had many seminiferous tubules. The germinal cells were lying near the basement membrane and showed stages of spermatogenesis. The supporting sertoli cells were seen in-between the gonial cells . The basement membrane of the tubules were normal. The lamina propria was seen out side the basement membrane. The interstitial stroma containing the Leydig cells. (Picture1). Even after three hours of noise exposure, the normal histological pattern of testis was maintained.

4.4 Histopathological changes in testis after chronic noise stress

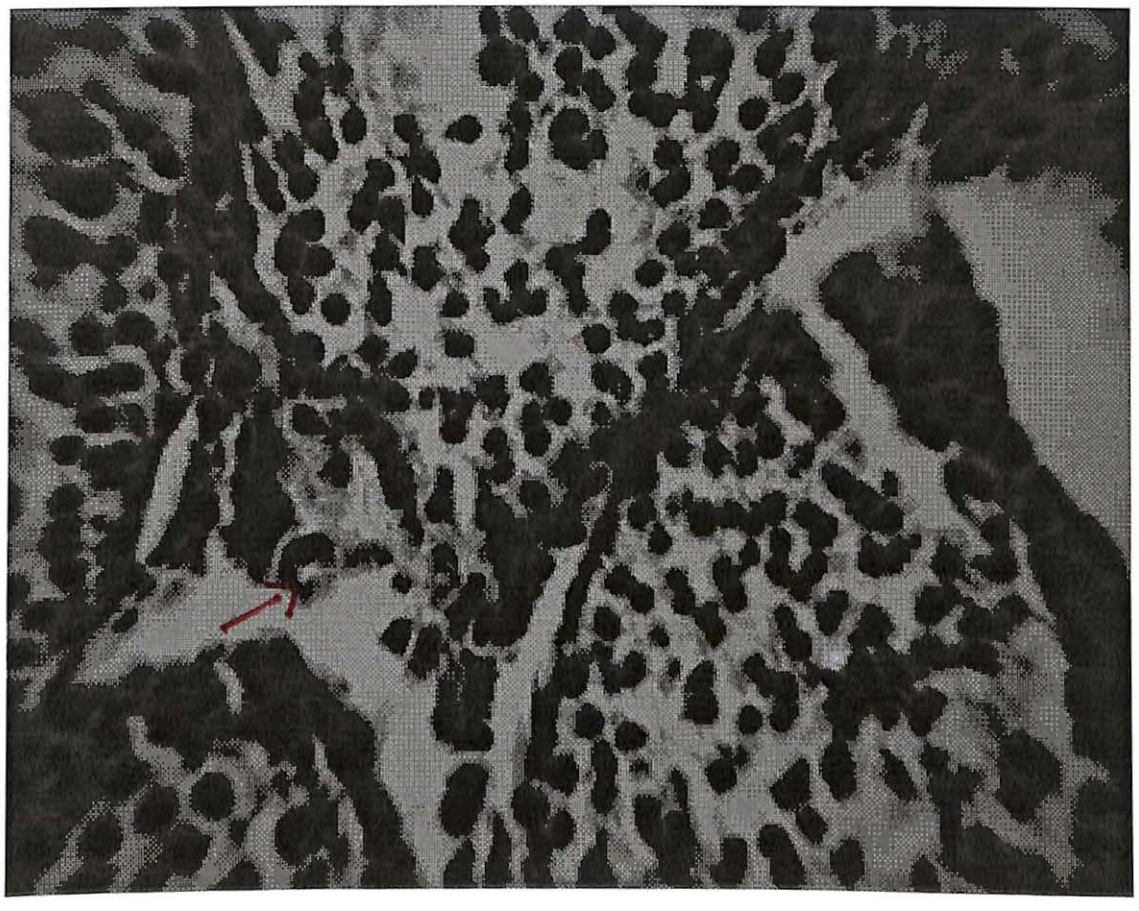
After 60 days of chronic noise exposure in one hour and three hours

duration there were changes noted in the arrangement of lobes and lobules. The centrally placed lobes were shrunk and separated from each other but normal arrangements were maintained in the periphery. There were maturation arrest in the stages of spermatogenesis noticed in some of the tubules. (Picture 2).

Even after 90 days of exposure to noise for one hour and three hours , the same picture was maintained as in 60 days. But some tubules showed interruption in the basement membrane.. The basement membrane was thinned out in some tubules. Degenerative changes were noted in some tubules. There were maturation arrest seen in few seminiferous tubules (Picture 3). These changes were very prominent compared to control groups. (PICTURE 4)

dscn1394.jpg (1280x960x24b jpeg)
90DAYS 3HRX40 Maturation arrest

PICTURE 1



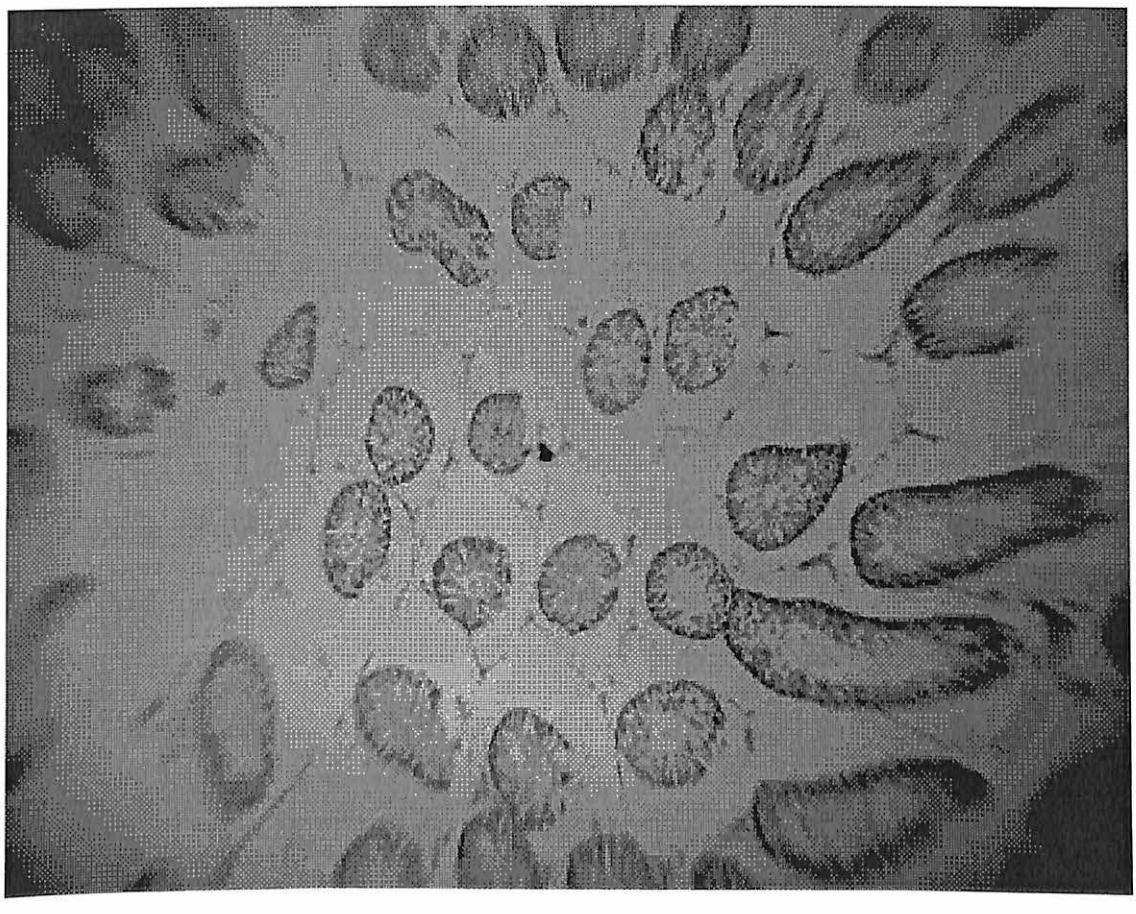
TESTIS 60DAYS3HR,X100 LEYDIG CELLS

PICTURE 2

TESTIS 90DAYS3HR,X40,MATURATION ARREST IN TUBULES

dscn1446.jpg (640x480x24b jpeg)
90days3hr ,x10 testicular lobes are far apart

PICTURE 3



TESTIS 90DAYS-3HR,X4—TESTICULAR LOBES ARE FAR APART