

EFFECT OF HE-NE LASER ON BLOOD SERUM TESTOSTERONE AND TESTICULAR TISSUE IN ADULT MALE RATS

Nazhad Hussein Qader, Nadem Hana Meikha and Basheer Mohamad Ali

Animal Resource Department, College of Agriculture, University of Salahhadin, Kurdistan Region - Iraq.

(Accepted for publication: May 2, 2016)

Abstract:

The current study was conducted to examine the effect of He-Ne laser therapy on the blood serum testosterone level and testicular tissue in adult male rats. Thirty five Albino Western adult male rats aged 3-4 months and weighing approximately 250-300 g were used and divided in to three groups. The testicular tissue of rats in the first treatment was exposed to a dose of irradiation 1.02 j/cm² (40 second) once daily for three successively days, while second treatment was exposed to a dose of irradiation 2.03 j/cm² (80 second) once daily for three successive date, while the third group remained without any treatments (control).

The results showed that the process of irradiation adversely affected on the level of blood serum testosterone in the first and second treatment compared to the normal level in the control group. The histological examination in treatment one showed low reduction in numbers of sertoli , leydig and spermatid cells at day one, while in day two showed medium reduction in numbers of sertoli , leydig and spermatid cells, and high reduction in numbers of sertoli , leydig and spermatid cells in day three of irradiation. In treatment two, the results showed medium reduction in numbers of sertoli , leydig and spermatid cells at day one, while in day two showed high reduction in numbers of sertoli , leydig and spermatid cells and very high reduction in numbers of sertoli , leydig and spermatid cells in day three of irradiation . In conclusion the current study revealed that steers factor cause reduction in numbers of sertoli , leydig and spermatid cells lead to low fertility rate within increasing of duration and repetition of irradiation.

Keywords: - He-Ne laser, Rats, Testosterone

Introduction:

Laser has many biomedical applications since it can be used in medical fields, in bio-stimulation of various organs, at low energy level in speeding up the wound healing process and it has the ability to stimulate the formation of epithelial cells, and it reduces the inflammatory phase during the healing process (Meikha, 2005). The irradiation of diode laser 830 nm, at a dose 28.05 j/cm² in rats affected on both qualitative and quantitative changes of the epithelial cells in seminiferous tubules, and showed no effect on sertoli cells, yet there was an increment in spermatids number (Taha and Valojerdi , 2004). While the irradiation of He-Ne laser at a dose of 1 j/cm² and treating the rats with serotonin in their peritoneal cavity protect them from the negative effects of irradiation (Omran *et al.*, 2001). Another study on rats using Nd-Yag laser, the irradiation accelerated the spermatogenesis process, and a temporary reduction in testicular interstitial tissues (Huyan and Ren , 1986). The use of He-Ne laser on rams at three different doses (high, medium and low) showed that both medium and low doses have prolonged sperms vitality (Jiuming, 1989). The

aim of this study was to evaluate the effect of He-Ne laser irradiation (5mw) on the level of serum testosterone, and the testicular tissue of adult rats.

Materials and Methods:

This study was conducted at Salahaddin University - Erbil - College of Agriculture, Department of Animal Resource, from September 2011, until March 2012. In this study 35 male rats (western albino) aged 3-4months and weighed about 250-300 g. The rats were kept in cages at a temperature of 21°C, and exposed to a photoperiod of 12/12 hours (light/darkness), while free access to standard diet was in the form of pellets. The rats were kept in cages for 10 days prior to the beginning of the experiment, and randomly divided into three groups, each of the first and second groups included 15 rats, while the third (control) group included 5 rats. The testicular tissue of rats in the treatment one (T1) were irradiated once daily for three successively days to a dose of 1.02 j/cm² for (40 seconds), and treatment two (T2) were irradiated once daily for three successively days to a dose of 2.03 j/cm² for (80 seconds),

while the third group (control) remained without any treatment. After a period of 24 hours from each irradiation process in (T1 & T2) samples of blood serum and testicular tissue were collected for histopathological examination.

Histological biopsies were installed in neutral formalin solution. According to (Drury *et al.*, 1976), the formalin fixed samples were processed, sectioned and stained. The procedure was carried in the laboratory of Histopathology, Department of Pathology, College of Medicine, University of Salahhadin – Erbil. The thickness of the microtome sections ranged from 4-5

micrometers, while the prepared slides were stained with Hematoxylin and Eosine stain.

Results:

Serological Examination:

Results showed that the irradiation affect adversely on the level of serum testosterone in (T1&T2), which was decreased with repeated irradiation during three successive days, while the control group remained within the normal level (Table 1).

Table (1): Results of testosterone hormone level concentration in blood serum of radiated and control groups.

Irradiated Rats	T1	T2	Control
	Testosterone conc. in blood serum (ng/ml)	Testosterone conc. in blood serum (ng/ml)	Testosterone conc. in blood serum (ng/ml)
Day one	1.97	1.6	-
Day two	0.86	0.44	-
Day three	0.22	0.11	-
Without irradiation	-	-	3.54

Histopathological Examination:

Treatment One

The results indicated low reduction in the numbers of Sertoli , Leydig and Spermatid cells during the first day of exposing while medium reduction at day two, and high reduction at day three for all cells. As shown in the table (2) and figures (1-3).

Table (2): Results of Histopathological Examination of T1, T2 and Control Groups

Types of Cells	T1			T2			Control
	Day one	Day tow	Day three	Day one	Day tow	Day three	
Sertoli Cells	Low Reduction	Medium Reduction	High Reduction	Medium Reduction	High Reduction	V. High Reduction	Normal
Leydig Cells	Low Reduction	Medium Reduction	High Reduction	Medium Reduction	High Reduction	V. High Reduction	Normal
Spermatid Cells	Low Reduction	Medium Reduction	High Reduction	Medium Reduction	High Reduction	V. High Reduction	Normal

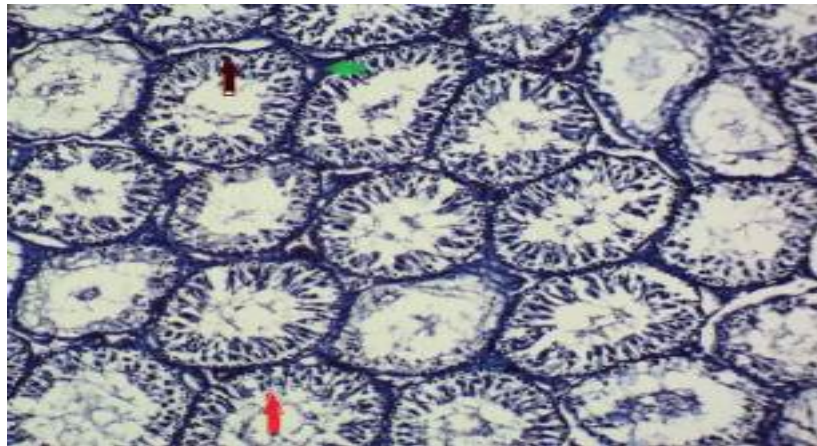


Figure (1): Shows histological changes of rats testis cells irradiated (T1) for day one. Low Reduction in sertoli cells (brown row) leydig cells (green row) and spermatids (red row), (H&E $\times 100$).

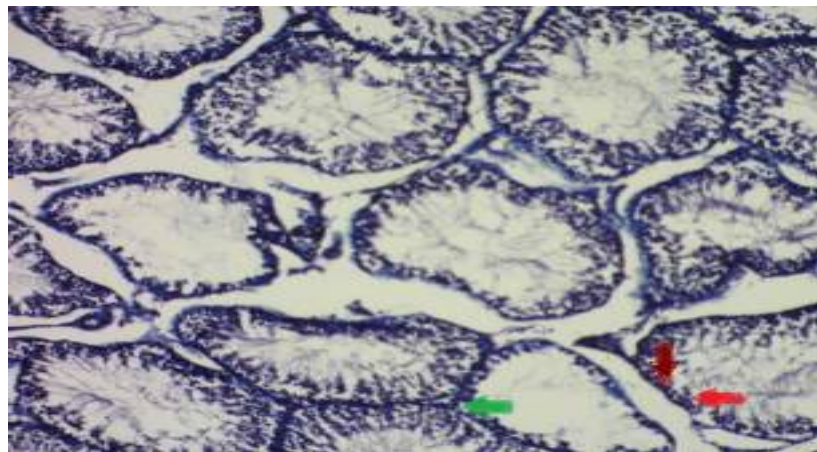


Figure (2): Shows histological changes of rats testis cells irradiated (T1) for day two. Medium Reduction in sertoli cells (brown row) leydig cells (green row) and spermatids (red row), (H&E $\times 100$).

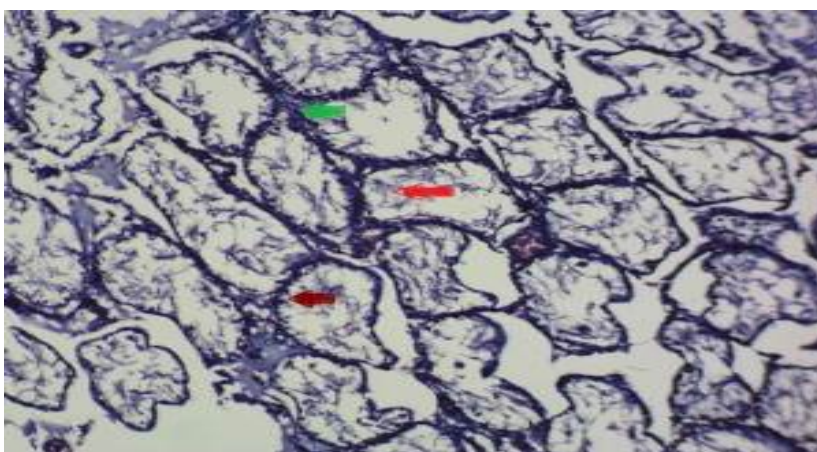


Figure (3): Shows histological changes of rats testis cells irradiated (T1) for day three. High Reduction in sertoli cells (brown row) leydig cells (green row) and spermatids (red row), (H&E $\times 100$).

Treatment Two

The results indicated medium reduction in the numbers of Sertoli , Leydig and Spermatid cells during the first day of exposure while high reduction at day two, and very high reduction at day three for all cells, as shown in the table (2) and figures (4-6).

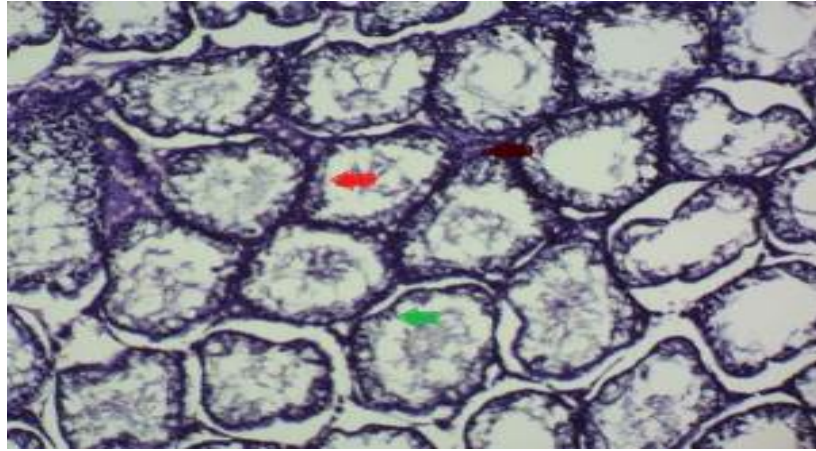


Figure (4): Shows histological changes of rats testis cells irradiated (T2) for day one. Medium Reduction in sertoli cells (brown row) leydig cells (green row) and spermatids (red row), (H&E \times 100).

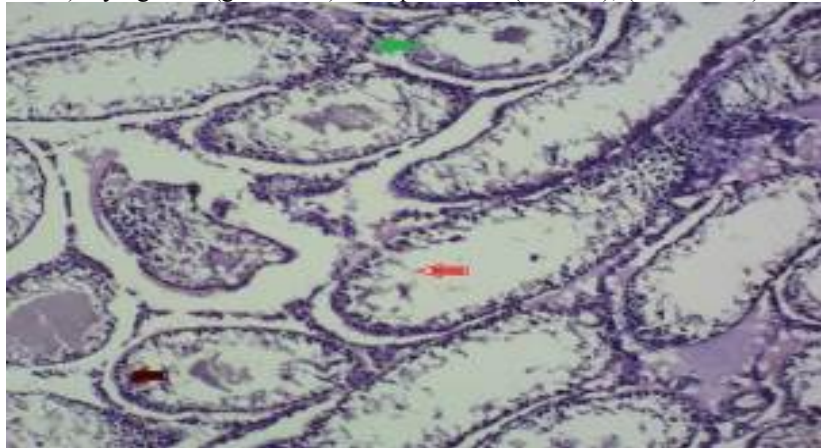


Figure (5): Shows histological changes of rats testis cells irradiated (T2) for day two. High Reduction in sertoli cells (brown row) leydig cells (green row) and spermatids (red row), (H&E \times 100).

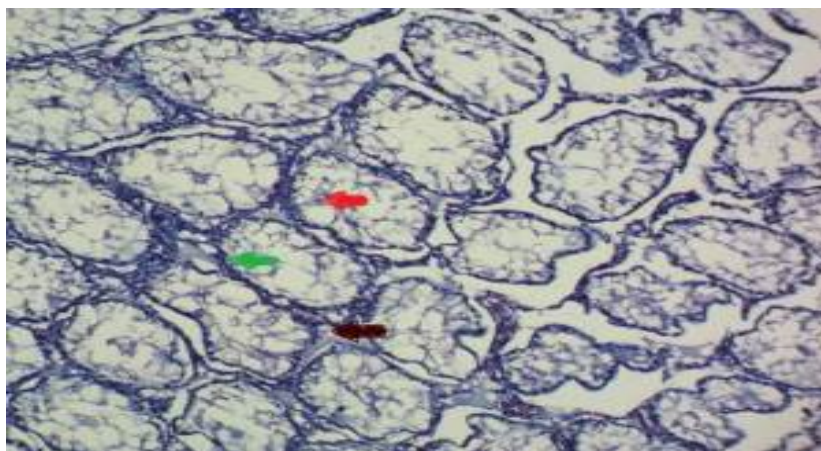


Figure (6): Shows histological changes of rats testis cells irradiated (T2) for day three. V. High Reduction in sertoli cells (brown row) leydig cells (green row) and spermatids (red row), (H&E \times 100).

Control group

Results showed a normal structure of testicular tissue. As shown in figure (7).

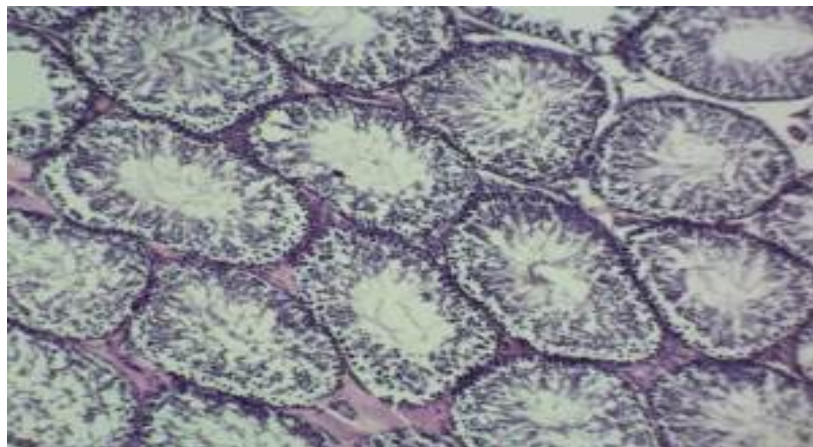


Figure (7): Shown the normal histological section of rats testis cells of (control group), (H&E \times 100).

Discussion:

Recently (Mailankto *et al.*, 2009) claimed that stress and pollution increase in our daily life due to the development in technology and the misuse of chemical materials, the study showed that rats exposed to mobile phone waves for a duration of one hour daily for a period of 28 days decreased semen quality and consequently fertility. The normal level of testosterone in the blood serum of male rats ranges from 3.51-3.96 ng/ml (Bartke *et al.*, 1973, Najim, 2012). Results of our study showed that the average testosterone concentration in adult male rats in the control group was 3.54 ng /ml. While in (T1&T2) showed reduction in the level of testosterone with the increasing of number and length of the period of irradiation. The results agree with those Omran *et al.*, (2001) who reported the adverse effect of He-Ne laser on rats blood serum testosterone concentration without using serotonin as a protective agent from laser irradiation, which acts in maintain the necessary cholesterol level to form testosterone in leydig cells. The natural formation of testosterone hormone in leydig cells is made by fats and interaction between (LH) and its receptors in leydig cells (Arthur *et al.*, 1998). Irradiation works on the inhibition of testosterone hormone formation process, and this might be explained by the effect of laser on (LH) receptors in testicular interstitial tissue, which proved by (Orr and Mann, 1992). The stress increase glucocorticosteroid hormone level that adversely affects receptors in leydig cells, which leads to

decrease in testosterone levels. (Knoll, 1991) pointed that stress generally affects on LH hypothalamo-pituitary- testis axis which leads to decreased in both (LH) and testosterone levels. While Irine *et al.*, (1980), showed a decrement in testosterone level as a result of stress which leading to an increase in deposited lipid droplets in leydig cells and consequently reduction of testosterone level.

Histological results in the present study showed that irradiation with He-Ne laser affected in both (T1&T2) adversely affected the numbers of leydig cells, and this disagree with the results of Irine *et al.*, (1980) who reported the stress had no effect on morphological and pathological changes of leydig cells during earlier stress days. On other hand histological results showed that the irradiation adversely affect the numbers of sertoli cells in (T1&T2) and cause increasing with the duration and repetition of irradiation .This result disagree with those reported by Taha and Valojerdi , (2004) who reported that the sertoli cells in rats testis were not affected by laser irradiation. Since male fertility and sperm formation process depend mainly on sertoli cells, therefore any damage to sertoli cells will adversely affect the process of spermatogenesis (William and Jing, 2005) that's agree with current study. The stress causes inhibition on spermatogenesis stages, by affecting hypothalamo-pituitary-testis axis, as well as fat accumulation in testicular interstitial tissue and thus there will be sever cellular degeneration (Rai *et al.*, 2003). Finally our study revealed that steers factor which cause reduction

in numbers of sertoli , leydig and spermatid cells lead to low fertility rate within increasing of duration and repetition of irradiation.

References:

- Al-Okaily Bara Najim (2012).protective effect of alcoholic extract of Black Current in Male Reproductive system of Methionine Overload Rats. The Iraqi J.Vet.Med.36(2):187-194.
- Arthur. G.H. Noaks .D.E. Pearson. H. and Parkinson.T.J (1998).Veterinary reproduction and obstetrics seventh Ed.W.B. Saunders company limited 553-559.
- Bartke, A. R.E. Steel, N. Musto and B.V. Calewell. (1973). Fluctuation in plasma testosterone levels in Adult Male Rats and mice .Endocrinology.92 (4): 1223-1228.
- Drury, R.A., Wallington, E.A. and Cameron, S.R (1976). Carletons histological technique , 4th ed. Oxford University press. London: 35,49-59.
- Huyan, Si-Le Ren-yu. (1986). Effects of irradiation to the rat scrotal testes on spermatogenesis with Nd:YAG laser. Chinese Journal of Biomedical Engineering- 04.
- Knoll BW (1991). Stress and the endocrine hypothalamus pituitary testis system. A review Vet.Q; 13 (2):104-14.
- Mailankot, M., Kunnath, A.B.,Tayalesksmih, Kodurn B. andValsalon, R. (2009). Radio frequency Electromagnetic Irradiation (free EMR) from gsm (0.9/1.8 ghz) Mobile Phones Induced Oxidative Stress Reduces Sperm Motility. Clinics, 64(6):561-565.
- Meikha, N.H. (2005). Stimulation Burn Healing Using 790 Diode laser in Rabbits. M.Sc. Thesis. Laser Institute for Higher Education. University of Baghdad.
- Omran, M.F., ABU-ZIED, N.M. and YACOB, S.F.(2001). Role of serotonin in the regulation of hormonal and antioxidant defense system in male rats exposed to laser radiation .National center for radiation research and technology, AEA, Cairo Egypt. (on line)
- Orr, T.E. and Mann, D.R. (1992) Role of Glucocorticoids in the stress induced suppression of testicular steroidogenesis in adult male rats. Hormone and Behavior; 26(3):350-63
- Pollard Irine ,BassettJR,and Joss M.P.Jean. (1980). Plasma testosterone levels and 3B-hydroxysteroid-dehydrogenase activity in the testis of the rat following prolonged exposure to stress. Journal of reproduction and fertility; 59: 101-106.
- Rai ,Jyoti;Pandey,S.N. and Srivasatava R.K. (2003). Effect of immobilization stress on spermatogenesis of albino Rats. J Ant .Soc.India 52 (1) 55-57.
- Taha, M.F. and Valojerdi, M.R. (2004). Quantitative and qualitative changes of the seminiferous epithelium induced by Ca. Al. As (830 nm) laser radiation. Lasers surgery and medicine. 34:352-359
- William H. Walker and Jing Cheng. (2005). FSH and testosterone signaling in sertoli cells Reproductive J1,130:15-28.
- Zhu Jiuming. (1989). Laser Irradiation Effect on Sperm Motility and Viability of Sheep. Application of Laser Issue.

كارىگهري تيشكى ليزه هيليويم – نيون بهسه ناستى هورموني نيرينه له مهسلى خوين له گهل شانهي گوني نيره جورجى بالغ.

پوخته:

نهم تويزينه وهيه نهنجام درا به مهبهستي تاقيكردنه وهى كارىگهري چارهسهري تيشكى ليزه هيليويم – نيون بهسه ناستى خهستي هورموني نيرينه له مهسلى خوين له گهل شانهي گوني نيره جورجى بالغ . سى و پينج جورجى نيري بالغ جورى ويسترن نهلباينو به تهمهني 3-4 مانگ به كيشى 250-300 گم به كار هات وه دابهش كرا بو سى گروپ .

گروپى يه كهم شانهي گون بهر تيشك كهوت به برى 1.02 جول/سم² (40 چركه) روژانه يهك جار بو سى روژى يهك بهدواى يهك .

گروپى دووهم شانهي گون بهر تيشك كهوت به برى 2.03 جول/سم² (80 چركه) روژانه يهك جار بو سى روژى يهك بهدواى يهك .

بهلام گروپى كونزوول (گروپى سييم) بهبى بهر كهوتنى تيشك مايه وه . له دهرئهنجامدا دهر كهوت كهوا تيشك كارىگهري نيگهتيفى هه بوو لهسه ناستى خهستي هورموني نيرينه له مهسلى خوين جورجه كاني بو ههردوو گروپى يه كهم و دووهم، به بهراورد له گهل گروپى كونزوول . كاتيگ دهرئهنجامى پشكيني شانهي له گروپى يه كهمدا دابهزىنى كهمى له ژمارهى خانه كاني سيستوتولى، له يدك وه سپيرماتيدي دهرخست له روژى يه كهم ، وه دابهزىنى مامناوهندي له ژمارهى خانه كاني سيستوتولى، له يدك وه سپيرماتيدي دهرخست له روژى دووهم . بهلام دابهزىنى بهرزى له ژمارهى خانه كاني سيستوتولى، له يدك وه سپيرماتيدي دهرخست له روژى سييم . له گروپى دووهمدا دابهزىنى مامناوهندي له ژمارهى خانه كاني سيستوتولى، له يدك وه سپيرماتيدي دهرخست له روژى يه كهم ، وه دابهزىنى بهرزى له ژمارهى خانه كاني سيستوتولى، له يدك وه سپيرماتيدي دهرخست له روژى دووهم . بهلام دابهزىنى زور بهرزى له ژمارهى خانه كاني سيستوتولى، له يدك وه سپيرماتيدي دهرخست له روژى سييم . له دهرئهنجامى نهم تويزينه وهيه دهر كهوت كارىگهري شه كهتتى هوكاره بو دابهزىنى ژمارهى خانه كاني سيستوتولى، له يدك وه سپيرماتيدي كه نهم شه هوكاره بو دابهزىنى ريژهى بيتين له نهنجامى زورى ماوه و دووباره كرده وهى تيشك .

تأثير أشعة الليزر – هليوم نيون على مستوى هرمون الشحمون الذكري في مصل الدم و النسيج الخصوي في ذكور الجرذان البالغة

الخلاصة :

اجريت هذه التجربة لأختبار تأثير أشعة الليزر – هليوم نيون على النسيج الخصوي و مستوى هرمون الشحمون الذكري في مصل دم الجرذان البالغة . استخدمت في هذه الدراسة خمسة و ثلاثون جرذا ذكرا بالغا من نوع ويسترن ألبينو بأعمار تتراوح ما بين 3-4 أشهر و أوزان 250-300 غم و قسمت الى ثلاثة مجاميع .

المجموعة الاولى تم تعريض النسيج الخصوي الى الاشعاع بجرعة 1.02 جول/سم² لمدة (40 ثانية) مرة واحدة يوميا و لمدة ثلاثة أيام متتالية . المجموعة الثانية فقد عرضت الى الاشعاع و بجرعة 2.03 جول/سم² لمدة (80 ثانية) مرة واحدة يوميا و لمدة ثلاثة أيام متتالية . في حين ان مجموعة السيطرة (المجموعة الثالثة) فقد تركت بدون معالجة اشعاعية . أظهرت النتائج بان عملية الاشعاع أثرت بشكل سلبي على تركيز مستوى هرمون الشحمون الذكري في مصل الجرذان للمجموعة الاولى و الثانية مقارنة بالمستوى الطبيعي لمجموعة السيطرة . بينما أظهرت نتائج الفحص النسيجي في المجموعة الاولى ان التشعيع أدى الى انخفاض طفيف في أعداد خلايا كل من سيرتولي ، ليدك و سليفات النطف في اليوم الاول من المعالجة . اما في اليوم الثاني كان الانخفاض متوسط في أعداد الخلايا الانفة الذكر . في حين ان زيادة التعرض في اليوم الثالث أدى الى انخفاض كبير في أعداد كل من كل من سيرتولي ، ليدك و سليفات النطف . اما المجموعة الثانية أدى التشعيع الى انخفاض متوسط في أعداد خلايا كل من سيرتولي ، ليدك و سليفات النطف في اليوم الاول من المعالجة . اما في اليوم الثاني كان الانخفاض عاليا في أعداد هذه الخلايا . في حين ان الانخفاض كان عاليا جدا في أعداد كل من كل من سيرتولي ، ليدك و سليفات النطف في اليوم الثالث . نستنتج من ما تقدم بأن التشعيع بالليزر – هليوم نيون يؤدي الى انخفاض في أعداد كل من خلايا سيرتولي، ليدك و سليفات النطف مما يؤدي الى انخفاض في معدل الخصوبة و يزداد هذا التأثير السلبي بزيادة الجرعة و تكرار فترة الاشعاع .