# Effect of Oxymetholone on The Sperm Quality and Sex Hormone Profile in Male Rats

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## Abstract

The objective of present study was to evaluated the effect of Oxymetholone drug on sperm parameters and sex hormone in adult male rats. Twenty male rats sexually mature were randomly divided into two groups; Each group has 10 rats. The treated group administered orally with Oxymethololne drug 5 mg/g body weight/ day of for 60 days while the control group administered orally normal saline per day. SPSS analysis of data generated with P< 0.05 considered statistically significant. The results showed a significant decrease ($p < 0.05$) in the mean sperm count, the motility and viability in treated group compared to control. Also, LH (luteinizing hormone) FSH (follicle stimulating hormone) and T (testosterone) of the treated group were significantly lowered compared to the control. The administration of Oxymethylene induces low sperm quality with reduction in sex hormone.

***Key words*** *: Oxymetholone, sperm parameters, hormonal profiles.*

## Introduction

Anabolic androgenic steroids (AAS) are among the most important compounds used in the medical field as well as used illegally by young athletes and non-athletes who use this stimulant to increase muscle mass and improve their functional performance [(1)] and AAS are synthetic derivatives of male sexual hormone testosterone and are small molecules that can spread negatively in various body tissues [(2).] The first effect is the anabolic effect or muscle building, which is an increase in protein synthesis, which leads to muscle growth and an increase in its size, while the second effect is the effect of ender, genetic or masculinity stimulant, including sperm production, roughness of the voice and frequent hair appearance; Although his pharmaceutical industry mainly possesses low-end and genetic effects called anabolic steroids, both effects are inseparable [(3).] Oxymethlone is a synthetic anabolic steroid that is structurally associated with male testosterone [(4,5 )] Uses Oxymethlone and other synthetic androgens to treat a variety of conditions, including hypogonadism and delayed puberty; androgens are also used to correct vascular edema, manage breast cancer, promote a positive balance of nitrogen after injury or surgery, and stimulate red blood cell production; athlete athletes consume large amounts of androgens in an effort to improve athletic performance [(5).] Hypothalamus -Pituitary Axis Hypothalamic- Gonodal (HPG) to regulate spermatogenesis; When the anterior pituitary gland's response to the stimulation of the hormones released to the canend (GnRH) decreases, the level of testosterone in the blood will rise, resulting in a decrease in FSH and LH secretion due to LH stimulation of testosterone production. If a specific cause, such as testicular damage, is caused by low levels of testosterone in the blood, resulting in an increased response of the anterior pituitary gland to stimulate the hormones releasing (GnRH), causing an increase in the secretion of LH and FSH hormones, thereby increasing the production of testosterone by the cells of Leydig Celle [(6)] and testosterone stimulates the maturation of spermatocyte [(7)]. Based on the above and for the purpose of knowing the effect of the sports stimulant Oxymetholone on

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reproductive hormones and sperm parameters, this study was designed to investigate the effect of the drug on the reproductive system of adult male rats.

## Materials and Methods

**1.Source of drug**

Oxymetholone was obtained in a dose (50mg) from a gym in Baghdad governorate. This dose is for humans and it is in pill form. Adult white male rats were used in this study using the concentration (5mg / kg)

**2. Animal:**

Three-month-old mature and healthy adult male rats weights ranging from (190-220 g) were procured from animal house of Faculty of Education for Girls-University of Kufa . They were maintained under controlled light schedule (12h Light:12h Dark) at room temperature (28 ºC) and with constant humidity ( 40 -50%). The animal acclimatized for a period of 7 days before the start of treatment . During this period, they were fed with standard rat chows/pellets and water.

**Experimental trial design procedure:**

**3.** The treatment group which included (10) adult male rats, as these animals were administrated orally with oxymetholone at a dose (5mg/kg) and by (1ml) for (60) days. The control group, which included (10) adult male rats, as these animals were administrated with normal saline (1ml) for 60 day

**4. Animal sacrifice**

Weights were recorded for all animals and then the animals were drugged by injecting them with the mixture of Xyline and ketamine, by taking (0.5ml) of ketamine and (0.1ml) of Xyline per (250g) of body weight. A T-shaped hole was made in the abdominal cavity and then blood samples were drawn from the heart directly by heart stab to obtain a blood sample of (2-5ml), after which the genitals (testes and epididymis) were raised. Removal of fatty tissue from around the organs to record their weights using a sensitive balance, while the tails of barnacles are used to study sperm characteristics. After that, the organs were placed in plastic containers containing formalin solution at a concentration of (10%), and then subjected to microscopic examination to study the tissues of these organs.

5.Preparation of serum samples:

After the animals were sacrificed, blood samples were collected and placed in gel tubes and left to clot in order to separate the blood components from the serum. Then these tubes were centrifuged at a rate of ( 3000rmp) for a period of (10) minutes. After the separation process is completed, the serum is transferred into a serum test tube and kept at a temperature of (-20) degrees Celsius until the results are read [8]. Hormones were measured using several Enzyme Linked Immunosorbent Assay (ELISA) which are known ways to measure the concentration of hormones, including follicle stimulating hormone, luteinizing hormone, testosterone and progesterone, and that several kit to stack hormones of the reproductive system in humans can be used to measure the level of hormones in the animal. Except for FSH, the modus operandi was followed by the information attached with the kit measurement kit.

**6- Sperm count:**

The sperm was collected from the right tail of the spray after sacrificing the animals and dissecting them, where the sperm was placed in (10ml) of the solution in a petri dish, after which we calculate the sperm by placing a drop on the glass slide and examining it under the optical microscope [9].

**Statistical analysis:**

The statistics of the results were analyzed using the SPSS statistical system for the purpose of comparing control and transaction totals using the t test.

## Results

**1- Sperm quality analysis:**

Results of sperm quality analysis are presented in the Table 1. The obtained data showed that the sperm concentration, motility and vitality in the treatment group that administered orally with Oxymethololne drug (5mg / kg) for to 60 days decreased in compared to the control group at each evaluated time ($P<0.05$).

**Table 1. The obtained data showed that the sperm concentration**

| Sperm Parameters | Number of animal | Treatment group M ±SD | Control group M ±SD |
|---|---|---|---|
| Average sperm concentration per mg testis (million / mg) | 10 | 24 ± 3.06 | 49 ± 2.67 |
| Average testicular sperm concentration (million / ml) | 10 | 33 ± 5.68 | 77 ± 7.29 |
| Average sperm concentration in epididymis delicacy (million / ml) | 10 | 25 ± 2.69 | 72 ± 6.2 |
| The percentage of live sperm in the epididymis | 10 | 30 ± 1.12 | 90 ± 7.49 |
| The percentage of abnormal sperms in the epididymis delicacy | 10 | 58 ± 9.42 | 17 ± 5.31 |
| The percentage of motile sperm | 10 | 20 ± 0.6 | 65 ± 8.22 |
| The degree of sperm activity | | 1 ± 0.08 | 2.5 ± 0.62 |

Table No. (1) shows the decrease in sperm parameters for the comparison group

Five rats per group

Significant difference * $p < 0.05$

**2- Hormones:**

The results of the statistical analysis showed that there were significantly differences ($p<0.01$) in testosterone concentrations, follicle stimulating hormone, luteinizing hormone and progesterone, with a significantly decrease ($p<0.01$) observed for the control group.

**Table No. (2) shows the decrease in hormones for the comparison group**

| Hormones (U/ml) | Number of animal | Treatment group M ±SD | Control group M ±SD |
|---|---|---|---|
| Testosterone | 10 | 2.7± 1.1 | 6.6± 3.6 |
| Progesterone | 10 | 0.7± 0.7 | 2.4± 0.5 |
| LH | 10 | 2.5± 1.8 | 6.8±1.9 |
| FSH | 10 | 2.6± 1.4 | 7.8± 1.4 |

Table No. (2) shows the decrease in hormones for the comparison group

Five rats per group

Significant difference * p <0.05

## Discussion

### 1-Changes in sperm standards:

The current study showed decrease that sperm concentration, motility and vitality in treatment group. The results of this study were in agreement with previous studies [10-12]. Naturally mature sperm production is the basis of male fertility [13] and the production of sperm and testosterone in the testicle is regulated mainly by FSH and LH, which have been released from the pituitary gland and are the main organizers of sperm formation [14], and the low level of reproductive club nutrients may have prevented the initiation of sperm formation in animals. Because the suppression of the nutrients of the canons prevents the signal responsible for the initiation and completion of the process of the formation of sperm during the natural maturation of the developing or developed sperm [15], the initiation of the process of the formation and maintenance of sperm naturally and qualitatively requires the presence of sufficient levels of hormone nutrients for the feeding and testosterone and usually the insufficient levels of these hormones are associated with severe abnormalities in the sperm and may lead to the state of sperm and weakness , In addition, the function of the ancillary gonads also depends on the presence of adequate levels of testosterone in the circulatory system [16,17]. It develops sperm cells, sperm cell differentiation and sperm protruding, and contributes to the generation of FSH receptors in sertoli cells [14]. Androgen also has an important role in the final stages of the formation of the sperm [18] It stimulates the transformation of the freshness of the round sperm to a long-range during the cycle of sperm formation, as well as the deficiency of androgens disrupts the process of freeing sperm [19] by changing the sperm contact points with sertoli cells, leading to the premature separation of the round sperm from the cells of sertoli [20].

### 2- Changes in hormones:

The current study showed a decrease in testosterone and hormonal FSH,LH this is consistent with previous studies [21,10]. It may be attributed to the simultaneous use of doses of anabolic androgens which leads to metabolism and change in the endocrine glands [22,23]. This may lead to a tendency to increase daily doses of external anabolic androgens which has caused a decrease in the secretion of internal androgens [24], as expected from the continued use of anabolic steroids. Testosterone reduction may be attributed to testicular size atrophy and lack of gonad hormones [10,11]. Low testosterone and FSH and LH hormone levels may be attributed to the fact that anabolic steroids administered from outside the body lead to adverse reactions to the secretion of the gonads, which may be due to the direct effect of hypothalamus to reduce gnRH secretion, which results in a decrease in the secretion of both FSH, LH and therefore the least biosynthesis of the release of testosterone hormones from testes [25]. In addition, androgenic steroids may produce oppressive effects on the testes and on the production of adrenal androgenic hormones [26,27]. It may be attributed to the fact that anabolic steroids increase the secretion of prolactin [28,29]. It may be because testosterone or its receptors may facilitate prolactin secretion and estrogen stimulates the release of prolactin from the anterior pituitary gland [30, 31].

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