

# Do Antioxidant Oils Help in Sustaining the Testicular Weight in Phenytoin Induced Toxicity in Rats?

Antioxidant Oils  
Help in  
Sustaining the  
Testicular  
Weight

Khalique-ur-Rehman<sup>1</sup>, Masood Ali<sup>2</sup>, Khalid Shehzad<sup>5</sup>, Hina Khan<sup>3</sup>, Humaira<sup>4</sup> and Raja Faisal<sup>3</sup>

## ABSTRACT

**Objective:** To evaluate the effective role of anti-oxidant oils in restoring the testicular weight in phenytoin toxicity in rats.

**Study Design:** Experimental Study

**Place and Duration of Study:** This study was conducted at the Al-Tibri Medical College and Hospital, Isra University Karachi Campus from October 2019 to March 2020.

**Materials and Methods:** Total 32 numbers of male albino rats were randomly selected from the animal house of the institute. The animals were divided into three groups Group A induced normal diet, B given intra peritoneal injection of phenytoin, C was given corn oil and D was given virgin coconut oil. The Body weight was evaluated at the initial day and at the final day of the study. At the completion of the study the bilateral testicular weight was evaluated by using a normal digital weight scale, and compare with normal group. The data was represented in the form of Mean and compare the Mean Body weight by applying simple 't' test and the testicular weight was evaluated by applying One-way ANOVA, and the level of significance was considered at  $P < 0.05$ .

**Results:** The study results show no significant difference was found on comparison of initial body weight with the final weight among the different groups. There was a significant difference found on comparison of bilateral testicular weight of phenytoin induced toxicity group of animals as compare to other, while in virgin coconut group the animals' testicular weight was well restoring in comparison with corn oil.

**Conclusion:** The study results show significant reduction of organ weight in experimental group as compare to control group, and the group of virgin coconut oil remarkably maintain the bilateral testicular weight as compare to other. Although the body weight was remains unchanged. Anti-oxidant properties of virgin coconut oil due to its biochemical composition strengthen its role against the free radicals and achieve their desirable effects on male fertility. Virgin coconut oil is easy to use and easily accessible for everyone.

**Key Words:** Phenytoin, Virgin Coconut Oil, Anti-oxidant

**Citation of article:** Rehman K, Ali M, Shehzad K, Khan H, Humaira, Faisal R. Do Antioxidant Oils Help in Sustaining the Testicular Weight in Phenytoin Induced Toxicity in Rats? Med Forum 2021;32(8):118-122.

## INTRODUCTION

Prolong usage of antiepileptic drugs among the patients of epilepsy and seizures are commonly found among the population. There are so many drugs listed as an antiepileptic drug, the most common drug found on the large numbers of fertility functions.<sup>1</sup>

<sup>1</sup>. Department of Anatomy, Chandka Medical College Shaheed Benazir Butto Medical University Larkana.

<sup>2</sup>. Department of Pharmacology / Anatomy<sup>3</sup> / Physiology<sup>4</sup>, Al-Tibri Medical College and Hospital Karachi.

<sup>5</sup>. Department of Anatomy, Liaquat College of Medicine and Dentistry. Karachi.

Correspondence: Dr. Hina Khan, Associate Professor of Anatomy, Al-Tibri Medical College and Hospital, Karachi.  
Contact No: 0346-3318553  
Email: drhinsalman@gmail.com

Received: March, 2021

Accepted: May, 2021

Printed: August, 2021

These drugs can faster the metabolic activity of the sex hormones and results in the decrease levels of androgen index. The study proven that the phenytoin alters the level of testosterone hormones by effecting the Leydig cells in the interstitial space, and in turn causes an inhibitory effect on spermatogenesis.<sup>2</sup> These antiepileptic drugs can produces a significant reduction sperm motility, velocity, and profound effects on male fertility and reduce the functional activities of reproductive organs in male.<sup>3</sup> Phenytoin reduces the plasma availability of testosterone hormones, and it may lead to effect the morphological and physiological parameters of the seminiferous tubules. In many of the studies the fertility rate was compromised and can alter the testicular weight and histomorphological changes among the spermatozoa.<sup>4</sup> The corn oil is mainly identifying by its biological name is Zea mays, and it's an essential vegetable oil that is commonly used worldwide. Its anti-oxidant activity can tag it beneficial oil for cooking. Daily usage of oil can reduce the oxidative stress from the body. Linoleic acid is the main composition as a component of poly-unsaturated fatty

acid.<sup>5</sup> Spermatozoa composed of higher numbers of poly un-saturated fatty acids, and oxidative stress can cause massive defragmentation of DNA and leads to infertility.<sup>6</sup> Biological name of virgin coconut oil is well known as Cocos nucifera. Coconut oil also contains myristic acid and palmitic acid, that are an essential anti-oxidant products used worldwide<sup>7</sup>. Myristic acid is mainly found in animal fat, coconut oil, butter fat, palm kernel oil and breast milk. It is also found in spermaceti, that are main component of whale sperm<sup>8</sup>. An average amount of Lauric acid is available in coconut oil, and all these elements having remarkable anti-oxidant impact on male fertility. Virgin coconut oil studies establishing its positive impact on the production of testosterone hormone, and boost up the sperm motility and enhance the physiological function of male reproductive organs.<sup>9</sup>

**MATERIALS AND METHODS**

After taken an ethical approval from the concerned authority of the institute. The Quasi-experimental study was done at Al-Tibri medical college and Hospital, from October 2019 to March 2020. The animals were taken from the animal house of respected institute, and the male albino rats were randomly taken of weight between 150-200 gms. The rats were kept in different cages with the maintenance of proper light and dark cycle. Total 32 numbers of male albino rats were divided into four groups on the basis of therapeutic study design. Each group having total eight number of rats labeled as:

**Group A:** Control Group, received an intra-peritoneal injection of 1-unit normal saline solution along with a normal daily diet.

**Group B:** A dose of Phenytoin 10mg/kg/body wt., intra-peritoneal once everyday

**Group C:** Received Virgin Coconut Oil (6.7ml) along with Phenytoin 10mg/kg/body wt., intra-peritoneal once everyday

**Group D:** Received Corn Oil (2.5ml) along with Phenytoin 10mg/kg/body wt., intra-peritoneal once everyday

The study was completed after 6 weeks and body weight of the animals were taken on day 1 and at the end of the study of each group by using a normal digital weight scale. After completion of the study, the animals were anesthetized with ethanol and then the incision was given to abdominal wall in sagittal section from xiphisternum to pubic symphysis then both testes were removed from the scrotum were weighed by using digital measuring scale. For microscopic parameters tissue was removed from the testis and preserved in 10% formalin. The Body weight and testicular weight of right and left site were presented in the form of Mean (standard deviation), and the data was evaluated by means of SPSS version 20.0. The Mean body weight was comparing with simple ‘t’ test and testicular weight

were compared by applying One-way ANOVA followed by post hoc Tuckey’s test among different therapeutic groups. The level of significance was kept at  $\leq 0.05$ .

**RESULTS**

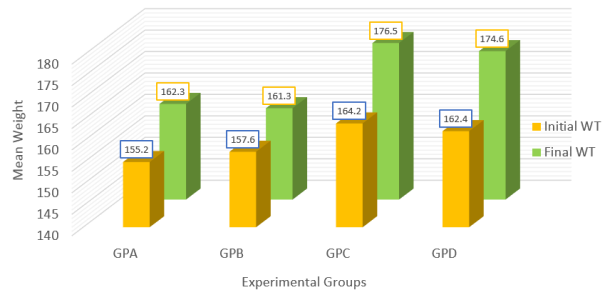
**Figure 1:** Shows the Comparison of Mean initial and final body weight of animals

**Table 1.a:** Shows the level of significance on comparison of Mean body weight among the different therapeutic groups

**Figure 2:** Shows the Mean Right sided testicular weight among different therapeutic groups.

**Figure 3:** Shows the Mean Left sided testicular weight among different therapeutic groups

**Table 1b:** Shows the level of significance on comparison of Bilateral Mean testicular weight among the different therapeutic groups.

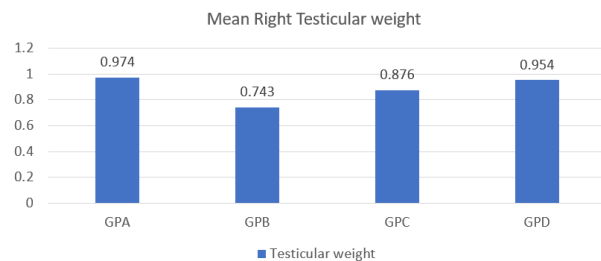


**Figure 1:** shows the Comparison of Mean initial and final body weight of animals

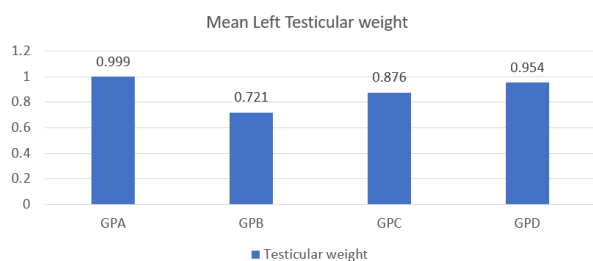
**Table No.1:** Shows the level of significance on comparison of Mean body weight among the different therapeutic groups

Groups	Compare the Mean of Initial and Final Body weight(gm)
A	$\leq 0.001$
B	$\leq 0.001$
C	$\leq 0.001$
D	$\leq 0.001$

$P \leq 0.05$  Simple ‘t’ test was applied



**Figure No.2:** Shows the Mean Right sided testicular weight among different therapeutic groups



**Figure No.3:** Shows the Mean Left sided testicular weight among different therapeutic groups

**Table No.1b:** Shows the level of significance on comparison of Bilateral Mean testicular weight among the different therapeutic groups

Groups	Compare the Mean of Right sided Testicular weight(gm)	Compare the Mean of Left sided Testicular weight(gm)
B vs A	≤0.001	≤0.001
B vs C	≤0.001	≤0.001
B vs D	0.001	0.001
A vs C	≤0.001	≤0.001
A vs D	0.547	0.621
C vs D	0.001	0.001

P= ≤0.05 One-way ANOVA followed by Post hoc Tuckey’s test

## DISCUSSION

The chronic use of phenytoin effect’s male reproductive function. One of the studies was taken in Nigeria regarding effects of chronic usage of phenytoin in male reproductive function in rats which showed that phenytoin reduced fertility in male’s rats.<sup>1</sup> In accordance with the results of one of the studies, including the effects of anti-epileptic drugs on the male fertility. The evidence was established the major effects on the male fertility in terms of reduction in testosterone hormones, and markedly interfere in the process of spermatogenesis. They study also revealed the decreased testicular weight as in the present study.<sup>9</sup> One of the randomized studies was done on 28 numbers of male rats with the phenytoin-levetiracetam adjunctive treatment plan. The objective of the study to evaluate their effects on male reproductive function, and the observation established that there were no significant effects on body weight, while the organ weight was reduced with the loss of other cellular function and effects the morphology of the spermatozoa and Sertoli cell. As in the present study, the body weight was not affected with the prolong use of phenytoin, although the toxic group shows significant

reduction of bilateral testes weight.<sup>10</sup> The other study showed the combination of different anticonvulsants drugs also has effect not only on cellular function and cell count it is also significantly suppressed the testosterone levels and decreased the weight of testes, seminal vesicle and epididymis. Moreover, these drugs cause the cytoarchitecture of testicular tissue disorganization. Similarly, in the present study, the anticonvulsant drug effects and reduce the cellular function and testicular weight.<sup>11</sup> Over 60 days the administration of oral anti-epileptic drug in rats marked decreased the testicular weight, sperm cell concentration, percentage of live sperms with high motility and cause the elevation of percentage of abnormal spermatozoa. The chronic use of antiepileptic drug also effects the testicular morphology and histopathological changes. As in the present study the anticonvulsant drugs induced toxicity has found almost in every aspect of reproductive system.<sup>12</sup> The study is to evaluate the protective role of corn oil and virgin coconut oil in antiepileptic induced testicular toxicity. The antioxidant oils have tremendous antioxidant effect on male fertility. The coconut oil has antimicrobial agent to kill microorganisms. and corn oil has poly unsaturated fatty acids which is important for normal healthy spermatozoa. The fatty acids are help for fluidity of sperms and promote fertilization. The antioxidant oils protect the reproductive system and enhance the fertilization, as the present study the both antioxidant oils significantly protect the reproductive system and sustain the testicular weight.<sup>13</sup> Another study which has been taken on male albino rats to know the lycopene effect on testicular torsion which cause testicular injury or testicular ischemia. Lycopene is a red pigment (carotenoid) in fruits and vegetables with antioxidant properties. The albino rats were segregates in three groups, and after an induction of ischemia operated animals were treated with lycopene and corn oil by gavage. 5 mins after operation IR (Ischemia reperfusion) decreased sperm motility and count in both sides of testes and elevate abnormal sperm rate bilaterally and in IRL (Ischemia reperfusion with lycopene) there is decreased rate of abnormal sperm count, similarly as in the present study the virgin coconut and corn oil having the same composition of lycopene in their extracts efficiently reduces the abnormal cell count in both testes as well helps in maintaining the organ morphological and functional capabilities. The remarkable effects were shown by virgin coconut oil as compare to other.<sup>14</sup> The study has proven that the anti -oxidative oil reduced oxidative stress. The study executed to know the role of Methanolic extract of Tribulus terrestris fruit (METT) in another anti-epileptic drug induced testicular toxicity. In this randomly male rat were selected. Which are exposed to antiepileptic drug. Orally treated rats with METT have anti oxidative effect and causes

elevation in weight of testes and increase LH, FSH, sperm motility, sperm count and reduced degenerative changes. The present study is also revealing the significant anti-oxidants effects of oils to reduce the oxidation stress or sustain the drug toxication as well. The anti-oxidative therapy is retaining the testicular weight too.<sup>15</sup> In accordance with the study results of consumption of virgin coconut oil in alcohol induced testicular toxicity along with antiretrovirals therapy. The results were shown no significant difference in both testicular weight and body weight. Although in present study the similar oil was help in resurgence of testicular weight along with phenytoin induced therapy, and simultaneously had no harm to body weight.<sup>16</sup> In accordance with the evidences of the study-based results establishing the impact of lycopene (a basic biochemical component of coconut oil) in adriamycin-induced testicular toxicity showed a restoration in sperm count and the physiological function of the different sex hormones. Altogether they contribute to restore the organ weight and enhance the testicular function and their biochemical compositions. Likewise, in the present study the coconut oil boosts up the testicular functions and helps in restore the organ weight.<sup>17</sup>

## CONCLUSION

The study results show significant reduction of organ weight in experimental group as compare to control group, and the group of virgin coconut oil remarkably maintain the bilateral testicular weight as compare to other. Although the body weight was remains unchanged. Anti-oxidant properties of virgin coconut oil due to its biochemical composition strengthen its role against the free radicals and achieve their desirable effects on male fertility. Virgin coconut oil is easy to use and easily accessible for everyone.

### Author's Contribution:

Concept & Design of Study: Khalique-ur-Rehman  
Drafting: Masood Ali, Khalid Shehzad

Data Analysis: Hina Khan, Humaira, Raja Faisal

Revisiting Critically: Khalique-ur-Rehman, Masood Ali, Khalid Shehzad

Final Approval of version: Khalique-ur-Rehman

**Conflict of Interest:** The study has no conflict of interest to declare by any author.

## REFERENCES

1. Olutunde PF, Emmanuel OS, Moyosore SA, Olusola AA, Olutoyin OO, Ebenezer AA, Abiodun O, Olakunle JO. Chronic use of phenytoin reversibly suppresses fertility in male Sprague-

Dawley rats. *Scientific Research and Essays* 2010; 5(9):999-1004.

2. Bauer J, Blumenthal S, Reuber M, Stoffel-Wagner B. Epilepsy syndrome, focus location, and treatment choice affect testicular function in men with epilepsy. *Neurol* 2004;62(2):243-6.
3. Meng H, Zhang F, Gao X, Wang X, Li D, Cui X, Wang Z. Effects of phenytoin on structural aberration of human sperm chromosomes in vitro. *Zhonghua yi xue yi chuan xue za zhi= Zhonghua yixue yichuanxue zazhi. Chinese J Med Genetics* 2001;18(4):303-5.
4. Silva DA, Löbenberg R, Davies N. Are excipients inert? Phenytoin pharmaceutical investigations with new incompatibility insights. *J Pharm Pharmaceutical Sci* 2018;21(1):19-31.
5. Hwang O. Role of oxidative stress in Parkinson's disease. *Experimental Neurobiol* 2013;22(1):11-7.
6. Jimenez-Fernandez S, Gurpegui M, Diaz-Atienza F, Perez-Costillas L, Gerstenberg M, Correll CU. Oxidative Stress and Antioxidant Parameters in Patients with Major Depressive Disorder Compared to Healthy Controls before and after Antidepressant Treatment: Results from a Meta-Analysis. *J Clin Psychiatr* 2015;76(12):1658-67.
7. Nevin KG, Rajamohan T. Virgin coconut oil supplemented diet increases the antioxidant status in rats. *Food Chemistry* 2006;99(2):260-6.
8. Arunima S, Rajamohan T. Effect of virgin coconut oil enriched diet on the antioxidant status and paraoxonase 1 activity in ameliorating the oxidative stress in rats—a comparative study. *Food Function* 2013;4(9):140.
9. Goyal HO, Braden TD, Mansour M, Williams CS, Kamaleldin A, Srivastava KK. Diethylstilbestrol-treated adult rats with altered epididymal sperm numbers and sperm motility parameters, but without alterations in sperm production and sperm morphology. *Biol Reproduction* 2001;64(3): 927-34.
10. Carvalho RK, Santos ML, Souza MR, Rocha TL, Guimarães FS, Anselmo-Franci JA, et al. Chronic exposure to cannabidiol induces reproductive toxicity in male Swiss mice. *J Applied Toxicol* 2018;38(9):1215-23.
11. Osuntokun OS, Olayiwola G, Atere TG, Adekomi DA, Adedokun KI, Oladokun OO. Hypothalamic–pituitary–testicular axis derangement following chronic phenytoin–levetiracetam adjunctive treatment in male Wistar rats. *Andrologia* 2020;52(11):e13788.
12. Bairy L, Paul V, Rao Y. Reproductive toxicity of sodium valproate in male rats. *Ind J Pharmacol* 2010;42(2):90.

13. Faisal R. Protective Role of Antioxidant Oils in Phenytoin Induced Toxicity of Seminiferous Tubules in Rats. *Med Forum* 2021;32(4):79.
14. Hekimoglu A, Kurcer Z, Aral F, Baba F, Sahna E, Atessahin A. Lycopene, an antioxidant carotenoid, attenuates testicular injury caused by ischemia/reperfusion in rats. *Tohoku J Experimental Med* 2009;218(2):141-7.
15. Shalaby MA, Hammouda AA. Assessment of protective and anti-oxidant properties of *Tribulus terrestris* fruits against testicular toxicity in rats. *J Intercultural Ethnopharmacol* 2014;3(3):113.
16. Ogedengbe OO, Naidu EC, Akang EN, Offor U, Onanuga IO, Peter AI, et al. Virgin coconut oil extract mitigates testicular- induced toxicity of alcohol use in antiretroviral therapy. *Androl* 2018;6(4):616-26.
17. Ateşşahin A, Karahan İ, Yılmaz S, Çeribaşı AO, Bulmuş Ö. Lycopene prevents adriamycin-induced testicular toxicity in rats. *Fertility and Sterility* 2006;85:1216-22.