Original Article

Effect of Tranylcypromine on Spermatogenesis in Adult Male Rats

Mahboobeh Gholamzadeh [1], *Mehrdad Shariati* [*2], *Davood Moghadamnia* [3]

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ABSTRACT

Background: Tranylcypromine is a monoamine oxidase inhibitor. It inhibits the breakdown of dopamine, serotonin, epinephrine and other monoamines. The aim of this study was to investigate the adverse effects of trarylcypromine on the spermatogenesis in adult male rats.

Methods: In this study, 50 adults male Wistar rats, weighing 200-250g, were used and divided into 5 groups of 10. Control group that was not given the treatment; the sham group received normal saline as a solvent and the experimental groups 1, 2 and 3 received 10, 20 and 40 mg/kg of trarylcypromine orally for 21 days. At the end of the trial period, the testes of rats were removed, tissue sections were prepared, stained with hematoxylin and eosin, and the histological features were examined under light microscopy. The statistical analyses performed were ANOVA and Tukey tests, using SPSS software, version 19.

Results: The results demonstrated that the number of spermatogonials, primary spermatocytes, spermatids and Leydig cells in the experimental groups 2 and 3 receiving tranylcypromine significantly decreased compared to those in the control group. However, the number of Sertoli cells in the experimental groups did not change significantly at any dose of the drug compared to those in the control group ($P \leq 0.05$).

Conclusion: Tranylcypromine reduced spermatogenesis and the respective indices in rat testicular tissues.

Keywords: Male adult rats, Spermatogenesis, Tranylcypromine.

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INTRODUCTION

The pituitary-gonad is one of the most complex and active physiological axis in the living organisms. This hormonal axis controls the synthesis and secretion of androgens, sexual differentiation, the development of secondary sexual characteristics and the respective behaviors (1).

Tranylcypromine is a non-specific monoamine oxidase inhibitor (2). Studies have shown that tranylcypromine may have special benefits in the treatment of clinical depression in humans. This was supported by a recent positron emission tomography (PET) study on brain activity (3). However, treatment with tranylcypromine also requires a diet limited in tyramine (4).

Monoamine oxidase inhibitors (MAOIs) are mainly used in psychiatry to treat depressive disorders and in neurological diseases for the treatment of Parkinson's disease. While other nonspecific and irreversible classic MAOIs, such as phenelzine, cause hypertension when taken with tyramine (5,6), tranylcypromine leads to a severe hypotension, a decrease in REM sleep and dizziness (7).

Tranylcypromine is an irreversible monoamine oxidase inhibitor that has been used for mood disorders, such as resistant depression since the 1950s. Due to dietary constraints and fear of drug interactions, tranylcypromine has been replaced by modern antidepressants that lack the pharmacologic side effects (8). Recently, a study by Han *et al*. (9) demonstrated that tranylcypromine, a hydroxylcinnamic acid hybrid, has anti-tumor activity and may be a potential agent in the treatment of human cancers (9).

A study by Caraci *et al*. (10) in 2015 found that tranylcypromine had neuroprotective effects against beta-amyloid-induced neurotoxinemia. Heijnen *et al*. (11) showed that tranylcypromine is useful in the treatment of bipolar depression. In a more recent study (12), tranylcypromine reduced developmental damage and improved general hyperalgesia in mice with induced endometriosis. Another study by Yao (13) found that tranylcypromine may have therapeutic potentials for the management of Herpes simplex type 1 infection.

Due to the side effects of some of the chemical drugs, there has not been a complete investigation on the effects of tranylcypromine on spermatogenesis in humans. In this study, the effect of tranylcypromine has been investigated on testicular tissue. Our specific research questions focused on the effect of this drug on the rat spermatogenesis, and whether the effect is dose-dependent.

MATERIALS AND METHODS

Animals

We obtained 50 adult male rats, weighing 200-250 grams from the animal breeding section of Pasteur

1. Department of Biology, Shiraz Branch, Islamic Azad University. Shiraz, Iran.
2. Department of Biology, Kazerun Branch, Islamic Azad University. Kazerun, Iran.
3. PhD of Animal Physiology, Young Researchers and Elite Club, Shiraz Branch, Islamic Azad University. Shiraz, Iran.
*Corresponding Author: Mehrdad Shariati, E-mail: mehrdad.shariati412@gmail.com

Institute of Iran. The ambient temperature during the test was 20 ± 2°C throughout the day. All animals were exposed to 12 hours of standard light and 12 hours of darkness and had free access to water and food. At all stages of the tests, the ethics of working with laboratory animals, as approved by the Ministry of Health and Medical Education, was observed and followed.

Treatments

Rats were randomly divided into 5 groups of 10. The control group did not undergo any drug treatment. Sham group received only normal saline solution as a solvent. Experimental groups 1, 2 and 3 received 10, 20 and 40 mg/kg of tranylcypromine orally for 21 days, respectively (14).

Histological Investigations

After opening the rats' abdomen under anesthesia, both testicles were removed in all groups and were fixed in formalin buffer solution. After recording the weight, tissue sections were prepared and stained with hematoxylin and eosin stain (H & E). The next step involved investigation of changes in sperm density of seminiferous tubules and the number of interstitial and Sertoli cells, and the spermatogenesis features. In order to count the cells in each group, 40 cross sections of seminiferous tubules were selected and after cell identification, cell counts were performed. In each group, the mean values representing various cell counts were recorded, and followed by the statistical evaluation of the data (15).

Statistical Analysis

In order to analyze the data, we used SPSS program version 19 to perform ANOVA, *t*-test and Tukey tests, comparing the results among the groups, with the significance level set at $P \leq 0.05$.

RESULTS

The mean number of spermatogonial cells, primary spermatocytes, spermatid and Leydig cells in the experimental groups 2 and 3 that received tranylcypromine decreased significantly ($P \leq 0.05$) compared to those in the control group (Table 1). However, the mean number of Sertoli cells in the experimental groups at any dose of the drug did not show a significant change compared to the same cells in the control group (Table 1).

Table 1. The mean number of spermatogonials, primary spermatocytes, spermatids, Sertoli and Leydig cells.

Group	Spermatogonials	Spermatocytes	Spermatids	Sertoli Cells	Leydig Cells
Control	49±1.4	68.1±2.3	113±11.3	15.3±3.3	19.2±0.4
Sham	49.1±1.7	68.4±203	112.4±25.2	15.6±2.2	19.9±1.1
Experimental 1	45.8±19	64.8±2	106±23.2	15.6±1.2	16.8±1.2
Experimental 2	39.5±1.7*	58.1±3.1*	96±13.15*	15.5±3.4	15.5±0.6*
Experimental 3	32.1±1.8*	48.7±2.8*	85.6±24.8*	15.4±1.9	10.4±2.1*

* = Denotes a significant difference compared to data for the control group ($P \leq 0.05$).

Seminiferous Tubules and Epithelia in Control and Sham Groups

Seminiferous tubules in the control and sham groups (Figures 1 & 2) had normal walls with no change in the luminal spaces. In the epithelia of the seminiferous tubules, all spermatogenic cells were observed. The epithelia in the seminiferous tubules appeared thick and the interstitial tissue was less developed.

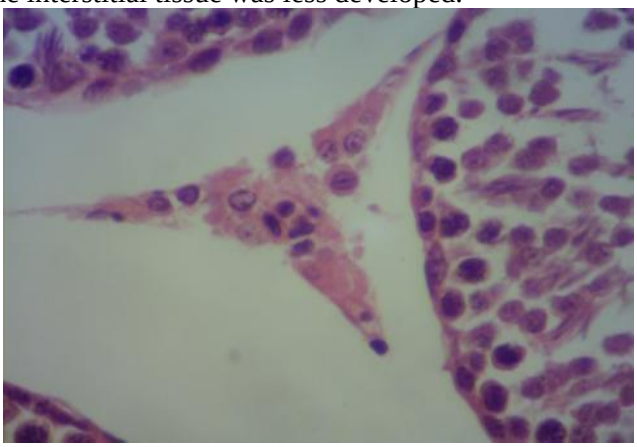

Figure 1. Photomicrograph of testicular tissue from control group (H & E stain; Mag. X40).

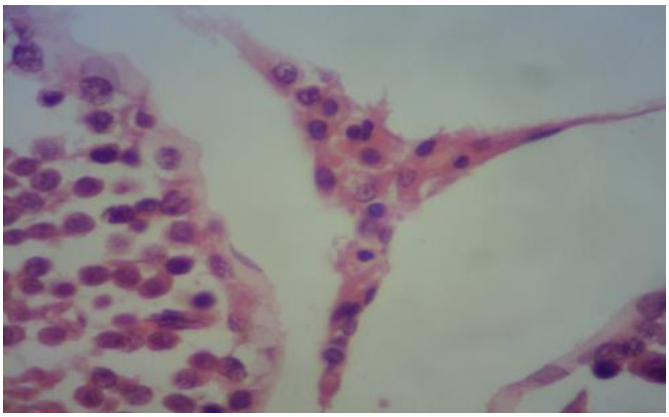

Figure 2. Photomicrograph of testicular tissue from sham group (H & E stain; Mag. X40).

Seminiferous Tubules and Epithelia in Experimental Groups

In experimental group 1 that received the minimum dose of tranylcypromine (10 mg/kg), a slight increase in luminal spaces was observed, the bulk of connective tissue among the tubules had grown, and sperms were present in the middle cavity of the tubules (Fig. 3). In experimental group 2, receiving a medium dose of the

drug (20 mg/kg), a large increase in luminal spaces was seen, the dimension of the epithelial cells had shrunk but the bulk of the connective tissue had grown. Also, the amount of sperms in the seminiferous tubules had decreased (Fig. 4). In experimental group 3, receiving the maximum dose of the drug (40 mg/kg), a large increase in the luminal spaces was noted, the dimension of the epithelial cells in the tubules had shrunk, and the amount of sperms in the tubules had decreased, but the bulk of the connective tissue in the tubules had increased (Fig. 5).

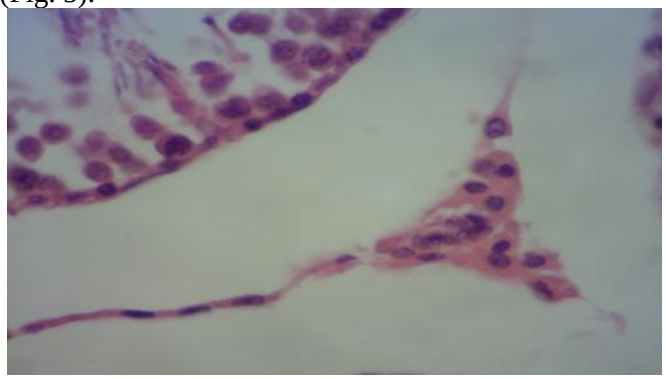

Figure 3. Photomicrograph of testicular tissue from the experimental group receiving 10 mg/kg tranylcypromine (H & E stain; Mag. X40).

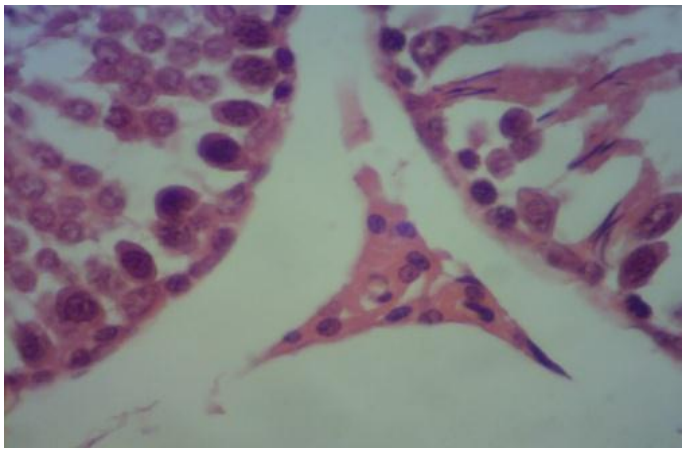

Figure 4. Photomicrograph of testicular tissue in the group receiving 20 mg/kg tranylcypromine (H & E stain; Mag. X40).

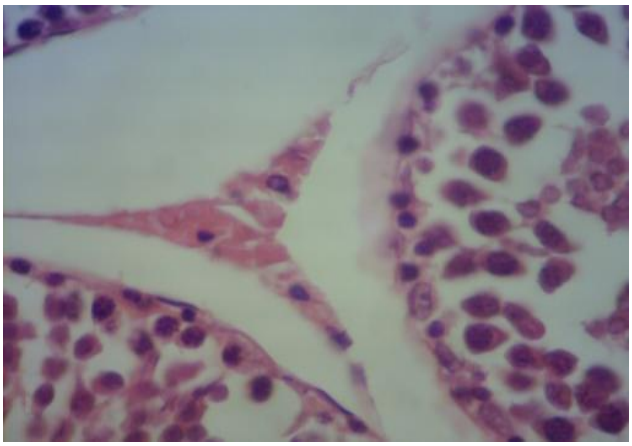

Figure 5. Photomicrograph of testicular tissue in the group receiving 40 mg/kg tranylcypromine (H & E stain; Mag. X40).

DISCUSSION

The results of this study demonstrated that the number of spermatogonials, primary spermatocytes, spermatid and Leydig cells in the experimental groups 2 and 3 significantly decreased compared to those in the control group. However, the number of Sertoli cells in the experimental groups, receiving tranylcypromine at any dose, did not significantly change compared to those in the control group.

Studies have shown that tranylcypromine acts as a monoamine oxidase inhibitor directly on the pituitary and decreases the secretion of luteinizing hormone (LH) in anterior pituitary gland. A decrease in LH may have effects on the activity of interstitial receptors in testicular tissue, leading to lower testosterone synthesis or release (16). The inhibitory effect of tranylcypromine on testicular development may be related to inhibiting the release of gonadotropins and blocking the environmental effects of LH. Also, tranylcypromine increases the concentration of dopamine in the brain striatum, thus its release via tuberoinfundibular dopaminergic neurons is transmitted to the portal circulation system, resulting in reduced prolactin secretion (17, 18). Lowering the secretion of prolactin appears to reduce the effects of LH on interstitial cells (16).

Procarbazine is a monoamine oxidase enzyme inhibitor similar to tranylcypromine (19). It has been shown that the short-term use of procarbazine improves spermatogenesis and fertility, but in the long-term suppresses the spermatogenesis and fertility (20). It has also been shown that treatment with procarbazine destroys approximately 70% of seminiferous tubules, reducing the secretion of testosterone and disrupts the spermatogenesis processes (21).

In a study by Urry *et al*. (22), it was found that after both short-term and long-term induction of a monoamine oxidase inhibitor, spermatogenesis and the growth of seminiferous tubules were inhibited (22). Studies also show that testosterone is the essential agent for the health and survival of spermatogenesis, and the development of spermatids, especially in the terminal stages (23). Consequently, reducing testosterone levels can disrupt spermatogenesis and may reduce the rate of spermatids production (23).

It is likely that the use of tranylcypromine exerts an inhibitory effect on the spermatogonial cell division, inhibiting the evolution and development of male sex cells, i.e., spermatozoids (24). It is also likely that increasing the dose of the drug promotes a further reduction in the production and/or development of spermatozoids (24).

The long-term use of tranylcypromine can be damaging and may lead to complete destruction of the testicular tissue. The reason may be that tranylcypromine affects the interstitial cells, thus destroying the spermatogenesis, rendering the person eventually infertile. Therefore, it is suggested that tranylcypromine should be used cautiously for clinical purposes. Due to the limited research being available on human subjects, future experimental studies to examine

the effects of tranylcypromine on the hypothalamus-pituitary-gonads axis in rats would provide valuable information. Also, conducting experimental studies on the effect of tranylcypromine on sexual behavior in rats and its role in the interactions among the hypothalamus-pituitary-gonads axis in female rats is recommended.

CONCLUSION

Based on the results of this study, oral use of tranylcypromine is likely to lead to structural changes in rat seminiferous tubules, causing disruptions in the spermatogenesis processes. Increasing the dose or the frequency of using this drug can also lead to further changes in the testicular organs and their cells. Our findings provided significant and distinct effects of tranylcypromine on the development and possibly fertility of adult male rats. The findings could provide a basis for conducting further comprehensive investigations on this drug in human subjects.

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CONFLICT OF INTEREST

There are no conflicts of interest in conducting this study.

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