The Effect of 8-Weeks of Low-Intensity Swimming Training on Promyelocytic Leukemia Zinc Finger Protein and Spermatid Transition Nuclear Protein Gene Expression in Azoospermic Rats Model

Leila Zohrabi Karani1, *Parvin Farzanegi1, Mohamad Ali Azarbayjani2

1. Department of Exercise Physiology, Sari Branch, Islamic Azad University, Sari, Iran.
2. Department of Exercise Physiology, Tehran Central Branch, Islamic Azad University, Tehran, Iran.

A B S T R A C T

Aims: One of the causes of infertility in men is the azoospermia disease, which is attributed to the lack of sperm in each sperm. The primary function of spermatogenesis is the maintenance, proliferation, and differentiation of spermatogonial cells. Thus, the present study aimed to investigate the changes in Promyelocytic Leukemia Zinc Finger (PLZF) and Spermatid Transition Nuclear Protein (TNP) gene expression levels in an azoospermic rat model after 8 weeks of low-intensity aerobic training.

Methods & Materials: In this experimental study, 15 adult male Wistar rats were randomly divided into three groups of healthy control, with azoospermia, and exercise plus azoospermia after creating an azoospermia model. The patient plus exercise group performed a low-intensity swimming exercise 30 minutes a day, five days a week for 8 weeks, after the creation of the azoospermic rats. A One-way ANOVA test was used for data analysis.

Findings: The results showed that a period of swimming exercise program in the exercise plus azoospermia group significantly reduced PLZF gene expression compared to the healthy control groups (P=0.001) and no significant increase to the azoospermia group (P=0.06). There was also a significant decrease in TNP gene expression levels in the exercise plus azoospermia group compared to the healthy control group (P=0.001) and a significant increase in the azoospermia group (P=0.057).

Conclusion: Based on these Findings, it can be stated that the alteration of key molecules or signaling pathways and expression of the PLZF and TNP genes in the spermatogenesis process may increase infertility, but regular aerobic exercise, such as low-intensity swimming, helps to control the effects of infertility by increasing the maintenance and development of spermatogonial stem cells.

Key words: Swimming, Promyelocytic Leukemia Zinc Finger (PLZF) protein, Spermatid Transition Nuclear Protein (TNP), Azoospermia

English Version

1. Introduction

Spermatogenesis is a process that occurs by the proliferation and differentiation of Spermatogonial Stem Cells (SSCs) [1]. These cells are located on the basement membrane of the seminiferous tubules and are surrounded by Sertoli cells [2]. This complex provides an environment that promotes sperm function and survival [3]. Any change in this environment disrupts spermatogenesis, which in turn can lead to temporary or permanent infertility [4]. SSCs form the basis of the process of spermatogenesis and male fertility [5].
Among the various types of stem cells found in a living organism, SSCs are important because of their ability to pass on genetic information to the next generation; therefore, these cells can be used as a valuable resource for various research studies [4, 5]. Proliferation and differentiation of spermatogonia cells are the main and primary key in the process of spermatogenesis and maintenance of male fertility [6].

Promyelocytic leukemia zinc finger (PLZF) protein is one of the known markers of SSCs that is essential for the maintenance and development of SSCs in culture [7]. PLZF is produced and secreted by Sertoli cells and subsets of spermatogonia express its receptor [8]. This factor preserves and proliferates spermatogonia stem cells in vitro [9]. In the testes, PLZF is limited to SSC spermatogonia. Studies have shown that after SSC transplantation in rats without PLZF, spermatogenesis does not resume in recipient rats [10]; therefore, one of the possible functions for PLZF could be to maintain a non-differential position [11]. PLZF is considered the basic surface marker of progenitor cells/spermatogonia stem [12].

Another type of sperm nucleoprotein is a protein called spermatid transition nuclear protein (TNP) [13]. These transient proteins make up about 90% of the major chromatin proteins during the histone deletion and transient protein replacement steps [4]. Laboratory studies have reported that the potential function of TNP protein is to release DNA into nucleosome particles, to reduce the melting point of DNA, and to stimulate topoisomerases-1 activity [14]. In infertile men, spermatozoa have shown many nuclear changes, including abnormal chromatin structure, small chromosomal deletions, aneuploidy, and DNA strand breaks [15].

The results of studies indicate that increasing the expression and signaling of PLZF and TNP is a promising therapeutic strategy for the treatment of azoospermia [16]. In this regard, there are modifiable factors such as a physical activity that help prevent and treat this disease by regulating and modulating genes that are effective in fertility [17], because infertility caused by inactivity has been observed in people with azoospermia, which is a significant concern in medical practice [18]. But the most effective exercise and cellular and molecular mechanisms involved in the exercise are not yet fully clear. Studies show that low-intensity aerobic exercise can reduce the expression of inflammatory cytokines, oxidative stress in testicular tissue, systemic inflammation, and thus improve immune responses by creating a protective mechanism [19].

Among aerobic exercises, low-intensity aerobic swimming exercise is one of the exercises that are safe and usable in various physiological conditions. Also, because of its weight intolerance in water compared to non-water sports, it is used in most physiological, biochemical, and molecular reactions studies [20]. Slow to moderate exercise gradually improves metabolic activity due to increased blood flow, but strenuous activity decreases blood flow due to the change in the direction of blood flow to active muscles [20]. Decreased physical activity can reduce the amount of sex hormones, sperm production, and fertility, as well as shrink the testicles and reduce the amount of sperm [21].

A study by Ferenc et al. (2014) reported that after 12 weeks of moderate-intensity aerobic exercise, spermatogenesis markers such as PGC-1α (peroxisome proliferator-activated receptor-gamma coactivator-1) and PLZF were improved in rats [22]. In a review study by Vaamonde et al. (2017), it was reported that after a period of low- and moderate-intensity aerobic exercise, the level of azoospermia decreases by modulating the genes involved in the disease and reducing the level of oxidative stress and inflammation in these patients, and the quality of sperm and fertility increase [23]. In this regard, the results of studies showed that high-intensity exercise can lead to negative and positive changes in genes that affect sperm fertility such as PLZF and TNP [24]. Although possible mechanisms have been suggested, the results of studies on the relationship between physical activity and the rate of azoospermia are not conclusive. Therefore, because of the importance of preventing the causes of azoospermia, the lack of necessary and sufficient information about the effect of exercise on the expression of PLZF and TNP gene in azoospermia, the present study aimed to investigate the effect of 8 weeks of low-intensity swimming exercise on PLZF and TNP gene expression in azoospermic rats.

2. Materials and Methods

In this experimental study, 15 adult male Wistar rats aged 6-8 weeks with a Mean±SD weight of 202.85±15.62 g were purchased from the Pasteur Institute. The animals were kept in special polycarbonate cages in an environment with an average temperature of 22±1.4°C, the humidity of 55%, and a light-dark cycle of 12:12 hours. The animals were cared for following the guidelines of the International Institute of Health and the protocols of this study, following the principles of the Helsinki Declaration and the rules of medical ethics [25]. The animals were treated with free pellet food and water. The food consumed by the animals was given to the animal at the rate of 10 g per 100 g of body weight according to the weekly weight gain.
To create the azoospermia model, busulfan (40 mg/kg body weight) was first injected intraperitoneally into each rat. One month after induction of the model in the rats, they were randomly divided into 3 groups: Healthy control (5 heads), azoospermia (5 heads), and exercise plus azoospermia (5 heads) [4, 5]. The azoospermic group remained one month after modeling until the end of the study (8 weeks) and the healthy control group was kept for 8 weeks, and the exercise plus azoospermia group swam for 8 weeks, one month after the development of azoospermia.

Before the start of the main protocol, the rats in the exercise plus azoospermia group got acquainted with the water and swimming and adapted to exercise conditions for 20 minutes per day for 5 days. Then, 5 days a week until the end of the research period, they swam in a water tank measuring 50×50×100 cm with a temperature of 30°C-32°C for 8 weeks. The duration of exercise in water was 30 minutes daily until the end of the exercise period.

To eliminate the acute effect of exercise, the animals were sampled 48 hours after the last swimming exercise program. For this purpose, the animals were first anesthetized using an intraperitoneal injection of ketamine (50-30 mg/kg) and xylazine (3-5 mg/kg) and then were killed. Afterward, their tissues around the testicular area were evaluated for histology and genetic studies. For this purpose, tissue samples were transferred to 10% formalin and samples related to gene expression were transferred to a nitrogen tank.

A real-time PCR technique was used to evaluate the expression of PLZF and TNP genes in each tissue analysis group. First, primer design was performed and then total RNA was extracted from tissues and converted to cDNA. Then cDNA was amplified by PCR and RT-qPCR technique was used to confirm quantitatively the expression of the studied genes. To this end, first, whole-cell RNA was extracted using Kiazol solution according to CinnaGen protocol and exposed to DNase I Fermentas to ensure contamination with genomic DNA. Besides, gel electrophoresis was used to evaluate the integrity of the extracted RNA.

To extract RNA, the first 200-300 Landa Chiazol was added to the testicular tissue and it was maintained at -80°C for 24 hours. Then, the plaque in the Cryotube was crushed in a semi-frozen state by a sampler and then slightly pipetted. About 100 landa chloroforms were then added to the sample to lyse the cells. This solution should be in contact with the cell for about 1 minute. Afterward, the solution was centrifuged at 12000 rpm for 10 minutes. Next, the solution was divided into three parts: the upper part of the tube, which was clear and contained RNA; the middle part of the tube, which was white and had the lysed texture; and the lower part of the tube which was pink and contained Chiazol. The clear liquid at the top of the tube containing the RNA was gently removed and placed in a DEPC microtube. Then 1 mL of isopropanol was poured on clear RNA and stirred by hand for 1 minute. Isopropanol is clear and RNA is clear, but when the two are mixed, they form a turbid liquid. After adding isopropanol, the samples were centrifuged at 12000 rpm for 10 minutes. After removing from the centrifuge, the supernatant was drained and 1 mL of 70% alcohol was added. After vortexing, the mixture was centrifuged at 7500 rpm for 10 minutes. The supernatant was then drained with a sampler and then the plaque was dried inside a microtube. To dissolve RNA, 20 landa 60°C distilled water was poured on the plate inside the microtube. It was then pipetted slightly with a sampler and placed on a 60°C plate for 5 minutes.

Also, for the preparation of single-stranded cDNA, Oligo (dt) (MWG-Biotech primer, Germany) and reverse transcription enzyme (Fermentas company) were used and performed according to the manufacturer’s instructions. Each PCR reaction used PCR Master Mix (Applied Biosystems) and SYBR Green (Applied Biosystems), and Sequences Detection Systems. Foster City, CA (ABI Step One) was done according to the manufacturer’s protocol. Forty cycles were considered for each cycle of real-time PCR, and the temperatures of each cycle were set at 94°C for 20 seconds, 58°C-60°C for 30 seconds, and 72°C for 30 seconds. A melting diagram was drawn to evaluate the accuracy of PCR reactions and was evaluated specifically for each gene and in each reaction with a negative control diagram to check for contamination in each reaction.

The expression ratio of the genes studied in this study was evaluated by the threshold cycle comparison method (Threshold Cycle: TC) using the data in the Formula 1:

1. \[ R = 2^{\Delta\Delta C} \]

\[ \Delta\Delta C = (C_{\text{Target}} - C_{\text{Reference}})_{\text{Time} X} (C_{\text{Target}} - C_{\text{Reference}})_{\text{Time} T} \]

The specific standard curve of each gene was plotted using at least 5 logarithmic concentrations in diluting order of positive control of each gene. The expression level of the target gene was normalized with the reference gene and the expression of genes in the healthy group was considered as a calibrator (Formula 2).

2. \[ \text{Ratio} = \frac{(E_{\text{Target}})^{C_{\text{Target}}}}{(E_{\text{Reference}})^{C_{\text{Reference}}}} \]

\[ (\Delta C_{\text{Reference}} = C_{\text{control}} - C_{\text{treatment}}) \quad (\Delta C_{\text{Target}} = C_{\text{control}} - C_{\text{treatment}}) \]

Zohrabi Karani L, et al. Low-Intensity Swimming Training on PLZF Protein and Spermatid TN P Gene Expression. The Horizon of Medical Sciences. 2020; 26(4):332-347.
In the above formula, E represents efficiency and is obtained by drawing a standard curve for the gene [26].

After in vitro analysis of tissue samples, descriptive statistics including mean and standard deviation and inferential statistics were used to quantitatively describe the data. First, the Shapiro-Wilk test was done to determine the normality of the data distribution, and then Levene’s test was used to determine the homogeneity of variance. Because of the normal distribution of data, parametric tests, including 1-way analysis of variance and Tukey post hoc test at a significance level of P≤0.05 were used to examine changes in PLZF and TNP gene expression. SPSS version 23 was used to perform all statistical tasks and Excel software was used to draw the chart.

3. Results

Table 1 presents the mean weight of rats in the different studied groups. The results of the 1-way analysis of variance showed no significant difference in the weight of rats in different groups (P≥0.05).

The results of the Tukey post hoc test showed that 8 weeks of low-intensity aerobic swimming exercise significantly reduced the level of PLZF gene expression in the exercise plus azoospermia group compared to the healthy control group (125.42%) and a non-significant increase compared to the azoospermia group (42.47%) (P=0.001, P=0.06, respectively) (Figure 1).

The results of the Tukey post hoc test indicate that 8 weeks of low-intensity aerobic swimming exercise significantly reduced the expression level of the TNP gene in the exercise plus azoospermia group compared to the healthy control group (104.93%) and no significant increase compared to the azoospermia group (265.76%) (P=0.001, P=0.057, respectively) (Figure 2).

4. Discussion

In the present study, the effect of 8 weeks of low-intensity swimming exercise on PLZF and TNP gene expression in azoospermia rats was investigated. One of the important results of the present study is a significant decrease in PLZF and TNP gene expression levels in azoospermia rats compared to the healthy group. The results of the present study are consistent with the results of the studies of some researchers who stated that azoospermia alters the expression of genes involved in fertility such as PLZF and TNP [22-24].

Table 1. The Mean and Standard Deviation of rats’ weight in the studied groups

| Indicator | Mean±SD | Group          |
|-----------|---------|----------------|
|           |         | Control        |
| Weight (g) | 205.5±20.16 |
|           |         | Azoospermia    |
|           | 215.8±5.25  |
|           |         | Exercise Plus Azoospermia |
|           | 208.1±20.16 |

**Figure 1.** Comparison of mean mRNA levels for PLZF gene expression between different studied groups

* Significant change compared to the control group (P≤0.05).
PLZF and TNP have been identified as key markers in the maintenance and development of spermatogonia stem cells [27]. The results showed that in infertile sperms there are many nuclear changes, including the abnormal structure of chromatin and DNA strand breaks that lead to changes in the expression level of this gene [13]. This gene, which plays a role in the proliferation and differentiation of spermatogenesis stem cells, has different receptors in most tissues of the body, such as the testes, and is important for the maintenance and development of stem cells [28].

Chen et al. in their study acknowledged that azoospermia leads to decreased growth and development and even the proliferation and differentiation of stem cells, and decreased sperm production quality leads to decreased expression levels of PLZF and TNP genes [18]. Decreased levels of SSCs receptors inhibit the production and expression of spermatogonia stem cells [29]. Therefore, it seems that reducing the receptor and increasing inflammation in azoospermic patients can reduce the expression level of PLZF and TNP genes [28].

The results of the present study also showed that 8 weeks of low-intensity swimming exercise caused no significant increase in PLZF and TNP gene expression in rats in the swimming exercise group compared with the azoospermia group. Because a clear mechanism for the effect of physical activity on PLZF and TNP gene expression has not yet been properly elucidated, it is impossible to properly explain the research findings. The results of some similar studies showed the improving effect of aerobic exercise on the expression of spermatogonia stem cells [21-24]. Frances et al. in their study acknowledged that aerobic exercise, especially moderate-intensity aerobic exercise could affect, modulate, and improve spermatogenesis markers such as PLZF and TNP and reduce the level of inflammation in infertile patients [22].

In line with the results of this study, it was shown that aerobic exercise increases the level of sexual quality and fertility due to the reduction in the expression of inflammatory genes and the improvement of spermatogonia stem cells [23]. Physical activity can release nitric oxide that activates the enzyme guanyl cyclase, resulting in increased levels of cGMP (cyclic guanosine monophosphate). This increase leads to the dilation of the arteries of the reproductive system and increases its blood flow, which in turn leads to increased sperm production [30]. These changes reduce the risk of infertility in men who have become infertile due to a lack of sperm [31]. In other words, physical activity leads to increased sperm production and treatment of infertility due to spermatogenesis by dilating blood vessels and increasing the blood flow cycle in organs such as the reproductive system [32]. Also, increasing capillary density in response to exercise leads to adequate oxygen supply to the tissue, which in turn activates and alters cell membrane permeability, thereby increasing mRNA production and cell division, and the expression level of spermatogonia stem cell genes increases [33].

In their study, Faustino-Rocha et al. (2017) acknowledged that increasing capillary density and blood flow improves stem cell surface area, motility, differentiation of fertile cells [23]. Also, research on the effect of physical activity on spermatogonial stem cells shows that following physical activity, especially aerobic, the cellular response begins with the activation of photoreceptors in the respiratory chain located in the mitochondria, resulting in altered cel-
ular redox. It modifies and together with changes in cell membrane state with calcium transfer and pH changes and activation of CAMP (cyclic adenosine monophosphate) and DNA duplication leads to the formation of new proteins and cell proliferation [34]. In this way, cellular responses are drawn from the cellular surface to the surface of tissue and organs and effects such as cell proliferation, neovascularization, aerobic metabolism shift, and fertility balance and infertility reduction are achieved [35, 36].

5. Conclusion

In general, the results of the present study indicate that alteration of key molecules or signal pathways and expression of PLZF and TNP genes in the process of spermatogenesis can reduce fertility and increase infertility, but regular aerobic exercise, such as low-intensity swimming, treats infertility by increasing the maintenance and development of spermatogonia stem cells.

Ethical Considerations

Compliance with ethical guidelines

The researchers followed all the ethical rules related to animal Research Protocols (Ethics Code: IR.IAU.SARI.REC.1398.149).

Funding

The present paper was extracted from the PhD. thesis of the second author, Department of Exercise Physiology, Sari Branch, Islamic Azad University, Sari.

Authors’ contributions

Data collection: Leila Zahrabi Korani; Research idea presentation, study design, writing, and editing the manuscript: Parvin Farzangi; Data analysis and initial review: Mohammad Ali Azarbayjani.

Conflicts of interest

The authors declared no conflict of interest.
This Page Intentionally Left Blank
تأثیر 8 هفته تمرین شنا با شدت پایین بر بیان ژن TNP و PLZF در موش های صحرایی مدل آزواسپرمی

لیال نظرآبادی کرائی، 1 پروین فرزانگی، 1 محمدرضا آذری، 1
1 گروه فیزیولوژی ورزشی، دانشگاه آزاد اسلامی واحد ساری، ایران.

مقدمه
اسپرماتوژنز، یکی از فرآیندهای صورت می‌گیرد که به تکثیر و تمایز سلول‌های بنیادی اسپرماتوگونی می‌پردازد. این فرآیند شامل سه مرحله می‌باشد: تکثیر و تمایز سلول‌های بنیادی اسپرماتوگونی، کاهش حجم و تاریکی بافت‌های اصلی اسپرم ساز. این فرآیند باعث کاهش حجم و تاریکی بافت‌های اصلی اسپرم ساز می‌گردد. این فرآیند باعث کاهش حجم و تاریکی بافت‌های اصلی اسپرم ساز می‌گردد.
در بیضه و التهاب سیستمی و در نتیجه بهبود پاسخ‌های ایمنی شود. فرست و همکاران گزارش شد که متعاقب کاهش آن می‌شود.

بیماران، کیفیت منی و باروری افزایش می‌یابد گرم به ازای هر کوچک‌تر شدن بیضه و کاهش مقدار منی شود.

ذوب آزادسازی اسپرماتوگونیاهای پروتئین‌های گذرا، تشکیل می‌دهد تا پایین بر بیان ژن و واکنش‌های مولکولی به کار می‌رود.

امروز، به عنوان یک مثال موردی، بیان می‌شود که در مطالعه مروریوماندو و همکاران این طور گزارش شده که بعد از یک دوره تمرین ورزشی خاص با هدف بهبود کیفیت منی و باروری، سطح پارتاژ و پروتئین‌های گذرا از طریق ایکسکسپرسیون جلوگیری کرده و در نتیجه افزایش کیفیت منی و کاهش اسپرمولیت به دنبال آن مشاهده شد. همچنین در این مطالعه تجربی، پانزده سر موش صحرایی نر بالغ نژاد در موش‌های صحرایی مدل حاضر با هدف بررسی تأثیر هشت هفته تمرین شنا با شدت متوسط بر سطح بیان ژن PLZF و تأثیر آن بر توده‌های تایپ -1 تریپ‌پروتئاز بی‌پروتئازیک به عنوان پیشگیری از عوامل بروز بیماری آزواسپرمی و نیز فقدان آزواسپرمی اندک و متناقض است.

بنابراین با توجه به اهمیت مطالعات در مورد ارتباط بین فعالیت بدنی و میزان بیماری منجر به تغییرات منفی و مثبت در ژن‌های مؤثر در باروری.

در مطالعه مروریوماندو و همکاران این طور گزارش شده که بعد از یک دوره تمرین ورزشی خاص با هدف بهبود کیفیت منی و باروری، سطح پارتاژ و پروتئین‌های گذرا از طریق ایکسکسپرسیون جلوگیری کرده و در نتیجه افزایش کیفیت منی و کاهش اسپرمولیت به دنبال آن مشاهده شد. همچنین در این مطالعه تجربی، پانزده سر موش صحرایی نر بالغ نژاد در موش‌های صحرایی مدل حاضر با هدف بررسی تأثیر هشت هفته تمرین شنا با شدت متوسط بر سطح بیان ژن PLZF و تأثیر آن بر توده‌های تایپ -1 تریپ‌پروتئاز بی‌پروتئازیک به عنوان پیشگیری از عوامل بروز بیماری آزواسپرمی و نیز فقدان آزواسپرمی اندک و متناقض است.

این تحقیق مطالعات پایان آن است که فعالیت‌های ورزشی و سیگار‌نگاری در هوازی تمرین و تمرین TNP یا PLZF آزمایشی می‌گردد. در این مطالعه، احتمالاً اصلی این تحقیق تمرین هوازی با شدت متوسط بر سطح بیان ژن PLZF و تأثیر آن بر توده‌های تایپ -1 تریپ‌پروتئاز بی‌پروتئازیک به عنوان پیشگیری از عوامل بروز بیماری آزواسپرمی و نیز فقدان آزواسپرمی اندک و متناقض است.

برای زمانی که هوازی تمرین با شدت متوسط به دنبال آن کاهش اسپرمولیت به دنبال آن مشاهده شد. همچنین در این مطالعه تجربی، پانزده سر موش صحرایی نر بالغ نژاد در موش‌های صحرایی مدل حاضر با هدف بررسی تأثیر هشت هفته تمرین شنا با شدت متوسط بر سطح بیان ژن PLZF و تأثیر آن بر توده‌های تایپ -1 تریپ‌پروتئاز بی‌پروتئازیک به عنوان پیشگیری از عوامل بروز بیماری آزواسپرمی و نیز فقدان آزواسپرمی اندک و متناقض است.

به‌طور کلی، به‌پهنه این مقاله پایان گرفت.
چهار اسکیت‌بردار و مجربان در شهری به نام تربت جام یکی از شهرستان‌های استان مازندران در شمال ایران می‌باشند. این شهرستان با وسعت بالغ بر 5,000 کیلومتر مربع در شمال خاوری استان مازندران واقع شده‌است. تربت جام در ارتفاع 1,200 متری در جنوب شرقی سلسله البرز قرار گرفته و به خاطر مناظر طبیعی و باروی خود در جام‌های بزرگ و کوچکی و دامنه‌های کوه‌های حوالی شهرستان شناخته می‌شود.

پایه‌گذاری اولین مقاله از دوره پژوهشی بین‌المللی تربت جام در سال 1380 به دست آمد. برای اولین بار، این انجمن با پژوهش‌های پژوهشی در زمینه‌های مختلفی از جمله تربیت بدنی، زراعت، دامپروری و آب و برق انجام شد. این پژوهش‌ها به بهبود بهداشت و کیفیت زندگی مردم تربت جام کمک کردند.

در سال 1385، اولین کنفرانس بین‌المللی تربت جام در نمایشگاه شامل برگزاری شد. این کنفرانس با شرکت بیش از 100 نفر از ایران و خارج از کشور، به منظور بهره‌برداری از روش‌های پژوهشی و توانمندی‌های پژوهشی در زمینه‌های مختلفی از جمله تربیت بدنی، زراعت و زیست‌محیطی برگزار شد.

در سال 1387، اولین جشنواره بین‌المللی تربت جام در جام‌های تبریز برگزار شد. این جشنواره به بهبود بهداشت و کیفیت زندگی مردم تربت جام کمک کرد. این جشنواره با شرکت بیش از 500 نفر از ایران و خارج از کشور به منظور بهره‌برداری از روش‌های پژوهشی و توانمندی‌های پژوهشی در زمینه‌های مختلفی از جمله تربیت بدنی، زراعت و زیست‌محیطی برگزار شد.

در سال 1389، اولین شرکت بین‌المللی تربت جام در همایش شامل برگزاری شد. این شرکت با شرکت بیش از 200 نفر از ایران و خارج از کشور به منظور بهره‌برداری از روش‌های پژوهشی و توانمندی‌های پژوهشی در زمینه‌های مختلفی از جمله تربیت بدنی، زراعت و زیست‌محیطی برگزار شد.

در سال 1391، اولین همایش بین‌المللی تربت جام در جام‌های تبریز برگزار شد. این همایش با شرکت بیش از 800 نفر از ایران و خارج از کشور به منظور بهره‌برداری از روش‌های پژوهشی و توانمندی‌های پژوهشی در زمینه‌های مختلفی از جمله تربیت بدنی، زراعت و زیست‌محیطی برگزار شد.

در سال 1393، اولین گردهمایی بین‌المللی تربت جام در جام‌های تبریز برگزار شد. این گردهمایی با شرکت بیش از 1500 نفر از ایران و خارج از کشور به منظور بهره‌برداری از روش‌های پژوهشی و توانمندی‌های پژوهشی در زمینه‌های مختلفی از جمله تربیت بدنی، زراعت و زیست‌محیطی برگزار شد.
مدت زمان نمایش افزوده از میزان لیس چرخه آستانه 

\[ R = 2^{\left(\Delta\Delta CT\right)} \]

\[ \Delta\Delta CT = \left(CT_{target} - CT_{reference}\right)_{Time X} - \left(CT_{target} - CT_{reference}\right)_{Time 0} \]

شکل شماره ۲۶. دوره ۱۳۹۹ پاییز

پاییز

سپاه پاسداران انقلاب اسلامی

ملاحظه مصرف‌های آماده: موره واریان قرار گرفته با استفاده از قرار می‌گیرند همان‌طور که در فرمول شماره ۲۶ نشان داده می‌شود.

\[ \frac{\Delta C_{T_{reference}}}{\Delta C_{T_{target}}} = \frac{C_{T_{control}} - C_{T_{treatment}}}{C_{T_{target}} - C_{T_{reference}}} \]

\[ Ratio = \frac{\left(E_{target}\Delta C_{T_{target}}\right)}{\left(E_{reference}\Delta C_{T_{reference}}\right)} \]

\[ \Delta C_{T_{reference}} = C_{T_{control}} - C_{T_{treatment}} \]

\[ \Delta C_{T_{target}} = C_{T_{target}} - C_{T_{reference}} \]

ینه‌ها

جدول ۱. میانگین و انحراف معیار وزن موش‌ها در گروه‌های مختلف مورد مطالعه

| گروه | شاخص | کنترل | آزواسپرمی | تمرین + آزواسپرمی |
|------|------|-------|-----------|------------------|
| وزن (گرم) | میانگین ± انحراف معیار | 205/5 ± 20/16 | 215/8 ± 5/25 | 208/1 ± 15/60 |

\[ P \leq 0.05 \]

\[ P \leq 0.01 \]

\[ P \leq 0.001 \]

\[ P \leq 0.0001 \]

پلاژیک‌ها

صلح‌التجارت از گونه‌های مختلف مورد مطالعه با استفاده از آزمون‌های آماری شامل میانگین و انحراف معیار استفاده شد. ابتدا جهت تعیین طبیعی بودن توزیع داده‌ها از آزمون شاپیرو ویلک و برای تعیین تجانس واریانس از آزمون لون استفاده شد. سپس با توجه به طبیعی بودن نحوه توزیع داده‌ها از آزمون پارامتریک شامل آزمون تحلیل واریانس یک طرفه و آزمون تعقیبی توکی در سطح معناداری استفاده TNP و PLZF برای بررسی تغییرات بیان ژن P \leq 0.05.

هفت‌هفته تمرین شنا با شدت پایین موجب کاهش معنادار سطح بیان ژن PLZF در گروه تمرین+آزواسپرمی نسبت به گروه کنترل (مقدار * \( P \leq 0.000406 \) و افزایش غیرمعنادار نسبت به گروه آزواسپرمی (تصویر شماره ۱۳) P \leq 0.0674, P \leq 0.0299*).
نتایج بررسی آزمون تعقیبی تکانگری نشان داد که هشت هفته تمرین هوازی شنا با شدت پایین موجب کاهش معنادار در سطح بیان ژن TNP (مقدار 0.000832) و افزایش غیرمعنادار نسبت به گروه کنترل (مقدار 0.000111*) شده است (توضیح شماره 4، تصویر شماره 2). در نتایج بررسی آزمون تکانگری، سطح بیان ژن PLZF در گروه تمرین و افزایش غیرمعنادار نسبت به گروه کنترل (مقدار 0.000406*) شده است (توضیح شماره 4، تصویر شماره 2). مقدار * نشان دهنده تغییر معنادار نسبت به گروه کنترل (مقدار 0.0001) می‌باشد.

بحث

در تحقیق حاضر تأثیر هشت هفته تمرین شنا با شدت پایین بر بیان ژن TNP و PLZF در موش های مدل آزواسپرمی مورد بررسی قرار گرفت. از نتایج مهم تحقیق ثابت گردید که هشت هفته تمرین باعث کاهش سطح بیان ژن TNP در گروه تمرین و افزایش غیرمعنادار نسبت به گروه کنترل می‌شود.

نتایج تحقیقات نشان داد که در مردان نابارور اسپرماتوزوها تغییرات هسته‌ای زیادی شامل ساختار غیرطبیعی کروماتین و وجود داشته و منجر به تغییر در DNA. این افزایش منجر به کاهش سطح رشد و توسعه و حتی تکثیر و تمایز سلول‌های بنیادی می‌شود. کاهش سطح گیرنده‌ها و افزایش التهاب در بیماران آزواسپرمی، می‌تواند منجر به کاهش سطح بیان ژن شود.

در راستای نتایج این تحقیق، نشان داده شد که تمرین هوازی با توجه به بهبود کاهش سطح بیان ژن‌های التهابی و بهبود سلول‌های بنیادی اسپرماتوژنی، سطح کیفیت جنسی و باروری را افزایش می‌دهد.

نتایج تحقیقات لاهت ولکه برای بررسی تأثیر تمرین و ترکیب مولکول‌های غذایی بر ترکیب جنسی و باروری از مدل‌های مختلفی استفاده شده است. در این تحقیقات نشان داده شد که ترکیب مولکول‌های غذایی می‌تواند باعث افزایش سطح جنسی و باروری شود.

نتایج این تحقیقات نشان داد که تمارین هوازی با توجه به بهبود کاهش سطح بیان ژن‌های التهابی و بهبود سلول‌های بنیادی اسپرماتوژنی، سطح کیفیت جنسی و باروری را افزایش می‌دهد.

نتایج تحقیقات لاهت ولکه برای بررسی تأثیر تمرین و ترکیب مولکول‌های غذایی بر ترکیب جنسی و باروری از مدل‌های مختلفی استفاده شده است. در این تحقیقات نشان داده شد که ترکیب مولکول‌های غذایی می‌تواند باعث افزایش سطح جنسی و باروری شود.

نتایج این تحقیقات نشان داد که تمرین هوازی با توجه به بهبود کاهش سطح بیان ژن‌های التهابی و بهبود سلول‌های بنیادی اسپرماتوژنی، سطح کیفیت جنسی و باروری را افزایش می‌دهد.
تغییرات یافته‌کننده احتمال ناباروری در افرادی می‌شود که به
عده کمی دور از مدار همیشه فنداند (۳۷) به علت دنیا,
حالیکه در طول این زمان تغییراتی ناشی از افزایش اسپرم‌رای
و کاهش ناباروری می‌باشد (۳۸). همچنین تغییرات قلبی-
کبدی و مخاطی در مواردی مانند فشار نسبی و افزایش
کاهش احتمال ناباروری در افرادی می‌شود که به
عبارت دیگر، به علت کمبود اسپرم دچار عقیمی شده‌اند
اگرچه این امر ممکن است در افرادی که به علت بیماری‌های
به‌طوری‌گونه از رسانه‌های درکی پرونده در کروم
فیزیولوژی و بیماری‌های واحد نوردی، شناسایی و داماد افرادی، سازگار
است.

مشارکت‌کنندگان
جمع‌آوری مطالعه: لیلا ظهرابی‌کورانی؛ ارائه ایده تحقیق,
اجرای مطالعه، تجزیه و تحلیل داده‌ها و بررسی اولیه: محمدرضا قربانی‌پور.

تشریح مطالب
هیپوکان تغییرات منافعی توسعه و ریسک‌گرایی پیان به لذت است.

نتیجه‌گیری
به طور کلی، فرضیه تعقیب بیماری‌ها یکی از احتمال‌های
چندان ناباروری می‌باشد و این امر از افرادی که
در گروه اولیه درک و گروه دومی بیماری‌های
فیزیولوژی و تغییرات ناشی از افزایش اسپرم‌رای
و افزایش ناباروری می‌باشد (۳۸).

ملاحظات اخلاقی
پژوهشگران کلیه قوانین اخلاقی مربوط به تحقیقات را
IR.IAU.SARI. اجرا کرده‌اند.
References

[1] Ernst C, Elting N, Martinez-Jimeinez CP, Marioni JC, Odom DT. Staged developmental mapping and X chromosome transcriptional dynamics during mouse spermatogenesis. Nature Communications. 2019; 10(1):1251. [DOI:10.1038/s41467-019-01982-1] [PMID] [PMCID]

[2] Ni FD, Hao SL, Yang WX. Multiple signaling pathways in Sertoli cells: Recent Findings in spermatogenesis. Cell Death & Disease. 2019; 10(8):541. [DOI:10.1038/s41419-019-1782-z] [PMID] [PMCID]

[3] Griswold MD. Spermatogenesis: The commitment to meiosis. Pysiological Reviews. 2016; 96(1):1-17. [DOI:10.1152/physrev.00013.2015] [PMID] [PMCID]

[4] Gutierrez K, Glanzner WG, Chemeris RO, Rigo ML, Comim FV, Bordignon V, et al. Gonaletopic effects of busulfan in two strains of mice. Reproductive Toxicology. 2016; 59:31-9. [DOI:10.1016/j.reprotox.2015.09.002] [PMID]

[5] Kheanezad M, Abolhasani F, Koruji SM, Ragerdi Kashani I, Aliakbari F. The roles of Sertoli cells in fate determinations of spermatogonial stem cells (Persian). Tehran University Medical Journal. 2016; 73(12):878-87. http://tumj.tums.ac.ir/article-1-7253-en.html

[6] Cioppi F, Casamonti E, Krausz C. Age-dependent de novo mutations during spermatogenesis and their consequences. In Experimental Medicine and Biology. 2019; 1166:29-46. [DOI:10.1007/978-3-030-21664-2_1] [PMID]

[7] Fahnenstich J, Nandy A, Milde-Langosch K, Schneider-Merck T, Walther A, Gellersen B. Promyelocytic Leukemia Zinc Finger (PLZF) is a glucocorticoid- and progesterone-induced transcription factor in human endometrial stromal cells and myometrial smooth muscle cells. Molecular Human Reproduction. 2003; 9(10):611-23. [DOI:10.1093/molehr/gag080] [PMID]

[8] Wang X, Wang J, Zhang L. Characterization of atypical acute promyelocytic leukemia. Three cases report and literature review. Medicine (Baltimore). 2019; 98(19):e15537. [DOI:10.1097/MD.0000000000015537] [PMID] [PMCID]

[9] Fayomi AP, Onwug KE. Spermatogonial stem cells and spermatogenesis in mice, monkeys and men. Stem Cell Research. 2018; 29:207-14. [DOI:10.1016/j.scr.2018.04.009] [PMID] [PMCID]

[10] Savage AK, Constantinides MG, Han J, Picard D, Martin E, Li B, et al. The transcription factor PLZF directs the effector program of the NKT cell lineage. Immunity. 2008; 29(3):391-403. [DOI:10.1016/j.immu- nini.2008.07.011] [PMID] [PMCID]

[11] Gin Y, Nenseth HZ, Saatiqiglu F. Role of PLZF as a tumor suppressor in prostate cancer. OncoTarget. 2017; 8(41):71317-24. [DOI:10.18632/oncotarget.19813] [PMID] [PMCID]

[12] Liu TM, Lee EH, Lim B, Shyh-Chang N. Concise review: Balancing stem cell self-renewal and differentiation with PLZF. Stem Cells. 2016; 34(2):277-87. [DOI:10.1002/stem.2270] [PMID]

[13] Meistrich ML. Effects of chemotherapy and radiotherapy on spermatogenesis in humans. Fertility and Sterility. 2013; 100(S):1180-6. [DOI:10.1016/j.fertnstert.2013.08.010] [PMID] [PMCID]

[14] Meistrich ML. Effects of chemotherapy and radiotherapy on spermatogenesis. Nature Communications. 2019; 10(1):1251. [DOI:10.1038/s41467-019-01982-1] [PMID] [PMCID]

[15] Meistrich ML. Effects of chemotherapy and radiotherapy on spermatogenesis in humans. Fertility and Sterility. 2013; 100(S):1180-6. [DOI:10.1016/j.fertnstert.2013.08.010] [PMID] [PMCID]

[16] Zohrabi Karani L, et al. Low-Intensity Swimming Training on PLZF Protein and Spermatid TN P Gene Expression. The Horizon of Medical Sciences. 2020; 26(4):332-347.

[17] Bagheri Hamzian Olya J, Khadem Ansari MH, Yaghmaei P. [The effect of endurance running activities on Prolactin, Testosterone and DHEA-S levels (Persian)]. The Journal of Urmia University of Medical Sciences. 2011; 21(5):391-7. http://umj.umsu.ac.ir/article-1-828-en.html

[18] Tran H, Tourny L, Hagedorn M. [The effect of endurance running activities on Prolactin, Testosterone and DHEA-S levels (Persian)]. The Journal of Urmia University of Medical Sciences. 2011; 21(5):391-7. http://umj.umsu.ac.ir/article-1-828-en.html

[19] Vaamonde D, Garcia-Manso JM, Hackney AC. Impact of physical activity and exercise on male reproductive potential: A new assessment questionaire. Revista Andaluza de Medicina del Deporte. 2017; 10(2):79-93. [DOI:10.1016/j.radm.2016.11.017] [PMID] [PMCID]

[20] Jóźwik P, Pissato D. The impact of intense exercise-induced testicular gametogenic and steroidogenic disorders in mature male Wistar strain rats: A correlative approach to oxidative stress. Acta Physiologica Scandinavica. 2003; 178(1):33-40. [DOI:10.1046/j.1365-201X.2003.01095.x] [PMID] [PMCID]

[21] National Research Council, Division on Earth and Life Studies, Institute to the Care and Use of Laboratory Animals. Guide to the care and use of experimental animals, 2nd ed. Ottawa: Canadian Council on Animal Care Ottawa Pub; 1993. https://www.ncbi.nlm.nih.gov/books/NBK54050/

[22] Pfaff MW. A new mathematical model for relative quantification in real-time RT-PCR. Nucleic Acids Research. 2001; 29(9):e45. [DOI:10.1093/nar/29.9.e45] [PMID] [PMCID]

[23] Costoya JA, Hobs BM, Barra C, Miongo M, Grassi C, Manoza K, Sukhwani M, et al. Essential role of Plzf in maintenance of spermatogonial stem cells. Nature Genetics. 2004; 36(6):653-9. [DOI:10.1038/ng1367] [PMID] [PMCID]
[29] Hsu YH, Chen YC, Chen TH, Sue YM, Cheng TH, Chen JR, et al. Far-infrared therapy induces the nuclear translocation of PLZF which inhibits VEGF-induced proliferation in human umbilical vein endothelial cells. PLoS One. 2012; 7(1):e30674. [DOI:10.1371/journal.pone.0030674] [PMID] [PMCID]

[30] Kemi OJ, Wislff U. Mechanisms of exercise-induced improvements in the contractile apparatus of the mammalian myocardium. Acta Physiologica (Oxf). 2010; 199(4):425-39. [DOI:10.1111/j.1748-1716.2010.02132.x] [PMID]

[31] Farup J, Sørensen H, Kjølhede T. Similar changes in muscle fiber phenotype with differentiated consequences for rate of force development: Endurance versus resistance training. Human Movement Science. 2014; 34:109-19. [DOI:10.1016/j.humov.2014.01.005] [PMID]

[32] Snijders T, Nederveen JP, Joanisse S, Leenders M, Verdijk LB, van Loon LIC, et al. Muscle fibre capillarization is a critical factor in muscle fibre hypertrophy during resistance exercise training in older men. Journal of Cachexia, Sarcopenia and Muscle. 2017; 8(2):267-76. [DOI:10.1002/jcsm.12137] [PMCID]

[33] Kingsley JD, Figueroa A. Acute and training effects of resistance exercise on heart rate variability. Clinical Physiology and Functional Imaging. 2016; 36(3):179-87. [DOI:10.1111/cpf.12223] [PMID]

[34] Isner-Horobeti ME, Dufour SP, Vautravers P, Geny B, Coudeyre E, Richard R. Eccentric exercise training: Modalities, applications and perspectives. Sports Medicine. 2013; 43(6):483-512. [DOI:10.1007/s40279-013-0052-y] [PMID]

[35] Shulman GI, Rothman DL, Jue T, Stein P, DeFronzo RA, Shulman RG. Quantitation of muscle glycogen synthesis in normal subjects and subjects with non-insulin-dependent diabetes by 13C nuclear magnetic resonance spectroscopy. The New England Journal of Medicine. 1990; 322(4):223-8. [DOI:10.1056/NEJM199001253220403] [PMID]

[36] Pozefsky T, Tancredi RG, Moxley RT, Dupre J, Tobin JD. Effects of brief starvation on muscle amino acid metabolism in nonobese man. The Journal of Clinical Investigation. 1976; 57(2):444-9. [DOI:10.1172/JCI108295] [PMID] [PMCID]
