

Research Article

Investigating the correlation between glutathione peroxidase and Interleukin-15 following aerobic exercise and probiotic supplementation in obese rats fed a high-fat diet

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Abstract

We investigate the effect of 6 weeks of aerobic training and probiotic supplementation on glycemic index and the correlation between glutathione peroxidase (GPX) and interleukin 15 (IL-15) in rats fed a high-fat diet. 40 male Wistar rats were divided into 5 groups (8 each): healthy control, high-fat diet (HFD), HFD+ aerobic exercise (Tr), HFD+ probiotic supplement (Sup) and HFD+Tr+Sup. Rats in the HFD group received a special high-fat diet for 2 months before the main protocol. The rats in the training groups ran on a rodent treadmill for 6 weeks, 5 days a week. Lactobacillus bifidus supplement was gavage to the rats of the supplemented groups in the amount of 10^9 CFU/ml after each round of training. The results of the statistical test showed that the HFD groups had a significant decrease in GPX compared to the healthy control group ($p < 0.05$). However, only the HFD+Tr+Sup group showed a significant increase in GPX compared to the HFD and HFD+Tr groups ($p < 0.05$). In contrast, HFD+Sup showed a significant increase in IL-15 gene expression compared to the healthy control group. Compared to the HFD group, the HFD+Tr and HFD+Tr+Sup groups showed a decrease and the HFD+Sup group showed a significant increase in IL-15 gene expression ($p < 0.05$). The correlation between GPX and IL-15 was not confirmed ($p < 0.05$). It seems that exercise training and lactobacillus supplementation are effective in regulating glucose, GPX and IL-15 indices after HFD. However, more studies should be done for the relationship and cross talk between antioxidant indices and IL-15.

Key Words: Correlation, Exercise training, Probiotics, High-fat diet, Interleukin-15

Introduction

Human metabolic diseases such as obesity, non-alcoholic steatohepatitis (NASH), and metabolic syndrome can be induced in rodents through genetic interventions and dietary modifications. Both strategies lead to increased body weight, hyperglycemia, hyperinsulinemia, hepatic steatosis, and heart failure (Buettnner et al., 2007). High-fat diets (HFD), consisting of 30% to 60% fat, include saturated fatty acids (SFA), monounsaturated fatty acids (MUFA), and polyunsaturated fatty acids (PUFA), and the consumption of such diets can last from a few days (short-term) to over a week (long-term) (Hulbert et al., 2005). Furthermore, insulin resistance, a critical step in the development of metabolic syndrome, can be influenced by the lipid content of the diet. Induction of insulin resistance occurs more frequently with diets rich in SFA and MUFA and may be minimized with PUFA consumption (Lee et al., 2006). Nutrient overload produced by HFD in laboratory animals and lack of physical activity are major risk factors for the development of metabolic syndrome in humans (James et al., 2012). When these conditions combine, they can exceed the capacity of adipose tissue to manage excess energy, resulting in increased levels of non-esterified fatty acids (NEFAs) and the release of pro-inflammatory cytokines and adipokines, which may lead to ectopic fat deposition in the liver, muscle, and heart (Després & Lemieux, 2006). In the liver, fat accumulation can be toxic (Schaffer, 2003). Liver tissue, by secreting inflammatory cytokines, can have autocrine and paracrine effects and influence distant tissues through the release of secreted substances into the bloodstream.

Interleukin-15 (IL-15) initially identified as a cytokine necessary for the maintenance of CD8⁺ memory T cells, has a multifaceted role in various tissues of the body (Waldmann & Tagaya, 1999). The liver hosts different subsets of lymphocytes, such as CD8⁺ T cells, NK cells, and NKT cells, which depend on IL-15 for their survival and maintenance (Crispe, 2009). IL-15 transcripts have been identified in various cell types of hematopoietic and non-hematopoietic origin in the liver. Hepatic resident macrophages, known as Kupffer cells are

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one of the main subsets that express IL-15 (Stienstra et al., 2010). The increase in this factor due to a high-fat diet results from the disruption of Kupffer cell homeostasis. Therefore, controlling this hepatic factor can have a positive impact on both liver tissue and other tissues in the body. Among the strategies for controlling cytokine-induced inflammation in the liver are regular exercise and the use of antioxidant and probiotic supplements.

Probiotics are currently used for the treatment or prevention of human diseases, conditions, and syndromes. They have also been shown to have a positive impact on neuroinflammation and pain, as well as on infections from seasonal diseases. Probiotics have a multifaceted effect, including a protective role in the gut. They compete with pathogens to produce direct antimicrobial effects and indirectly enhance intestinal barrier function (Ng et al., 2009). Many *Lactobacillus* species are used in food products. *Lactobacillus* species can improve conditions such as gastrointestinal diseases, allergies, and liver diseases through various mechanisms, such as producing metabolites that can directly inhibit pathogens (Hojsak et al., 2010). The microorganisms present in the gut also affect liver disease. This is because the portal venous system defines the gut-liver axis, and there is a close anatomical and functional interaction between the gastrointestinal tract and the liver (O'Hara et al., 2017). The beneficial effects of various probiotics on liver disease, both non-alcoholic and alcoholic, have been confirmed. However, their role in regulating hepatic interleukin-15 and serum antioxidants after a high-fat diet has not been investigated.

Regular exercise is one of the most effective non-pharmacological treatments for metabolic disorders associated with obesity and its complications. Exercise is widely used to control or treat obesity-related diseases (Church, 2011). Aerobic exercise induces intracellular adaptations in a specific manner across various tissues. Exercise has improved hyperglycemia, hyperinsulinemia, muscle glycogen loss, and atrophy in animals with liver damage (Barel et al., 2010). The beneficial effects of endurance exercise include increased insulin sensitivity and reduced fat mass. However, there is limited knowledge regarding the mechanisms by which endurance exercise exerts its beneficial effects. Myokines, cytokines secreted by skeletal muscles, have been recognized as potential mediators. Recently, the role of skeletal muscle-derived interleukin-15 (IL-15) in improving body composition and insulin sensitivity has been suggested. IL-15 may play a role in the systemic anti-obesity and insulin-sensitizing effects of endurance exercise, not only as a paracrine and autocrine factor but also as an endocrine agent. However, the hepatic effects of IL-15 in high-fat diets and its endocrine effects have not been investigated. Therefore, this study will examine the effects of exercise and probiotic supplementation on hepatic IL-15 and antioxidant effects in rats

with an obesity model.

Materials and Methods

Animals

In this experimental study, 40 male Wistar rats aged 10 to 12 weeks with an average weight of 240 grams were purchased from the Pasteur Institute of Iran (Tehran, Iran) and were maintained under standard laboratory conditions at the Animal Biology Laboratory of the Islamic Azad University, Science and Research Branch. The animals were acclimatized to laboratory conditions for two weeks under a 12-hour light/dark cycle, with an average temperature of 20 ± 22 degrees Celsius and an average humidity of 50 ± 5 percent, in ten standard rodent cages measuring $30 \times 15 \times 15$ centimeters, based on the laboratory animal care checklist of the Islamic Azad University, Science and Research Branch, Faculty of Medical Sciences and Technologies. During the first two weeks, the subjects were fed a normal diet consisting of pellets provided by the Javaneh Khorasan Company, containing 19.6% protein, 3% fat, 6% fiber, 50.4% carbohydrates, 13% moisture, and other components. This study was conducted under the supervision of the Animal Ethics Committee of the Islamic Azad University, Science and Research Branch, Tehran, Iran, with ethical code IR.IAU.SRB.REC.1402.188.

High-Fat Diet

Apart from the healthy control group, which was randomly selected, the remaining subjects were placed on a high-fat diet for two months, with the addition of 40% solid vegetable oil to their meals compared to a normal diet. The amount of added oil was considered to be 37% of the weight of the normal diet. The weight of the subjects was measured before the start of the diet and during the high-fat diet twice a week using a digital scale model BLK-500 by Sunli, with an accuracy of 0.1 grams.

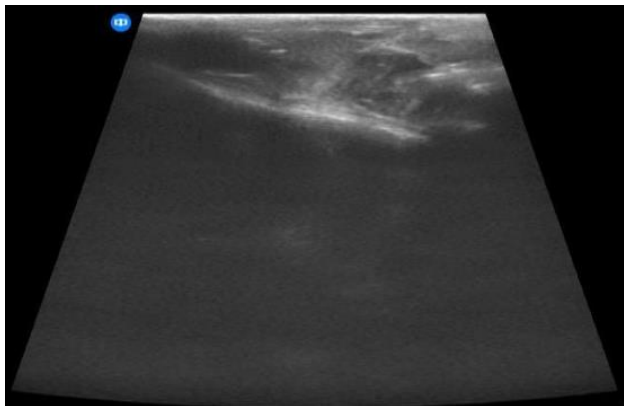
After two months, 32 subjects on the high-fat diet were randomly divided into four groups of eight, including the high-fat diet group (HFD), the HFD with exercise group (HFD+Tr), the HFD with supplementation group (HFD+Sup), and the HFD with supplementation and exercise group (HFD+Tr+Sup).

Before starting the exercise protocol and supplementation, four rats from the control group and 3 rats from the experimental group on a high-fat diet were randomly subjected to ultrasound examination of the liver and organs by a veterinarian. It was confirmed that, apart from one healthy control rat, the HFD subjects showed visible evidence of fatty liver (Figure 1).

Exercise

The HFD+Tr and HFD+Tr+Sup groups were subjected to exercise on a treadmill from the leading engineering company, Pishro Andisheh Sanat, model Navid Line, product code 12603,

A)



B)

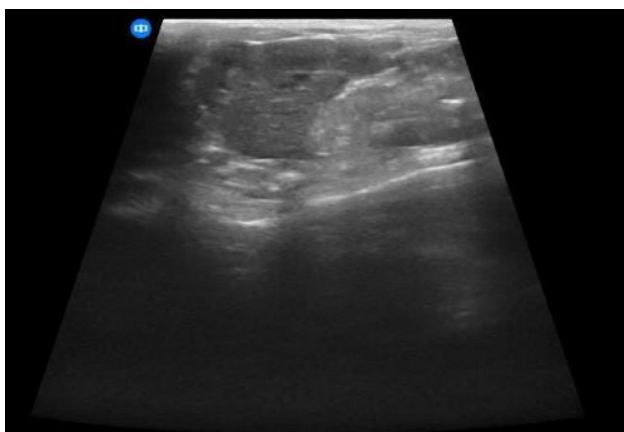


Figure 2. The image of ultrasound in healthy (A) and fatty liver group (B).

for 6 weeks, 5 days a week. The exercise method involved changes in intensity and duration as follows: Throughout the training period, a warm-up and cool-down phase of 5 minutes at a speed of 10 meters per minute was maintained at the beginning and end of each session. In the first week, the speed was set at 15 m/min for 30 minutes; in the second week, 18 m/min for 35 minutes; in the third week, 21 m/min for 40 minutes; in the fourth week, 23 m/min for 45 minutes; in the fifth week, 25 m/min for 50 minutes; and in the sixth week, 27 m/min for 60 minutes, implementing the exercise protocol (Karakioulaki et al., 2020).

Supplement

The HFD+Sup and HFD+Tr+Sup groups were supplemented with *Lactobacillus bifidus* obtained from the pharmaceutical company Zist Takhmir, registered under number 368211, containing a concentration of 109 CFU/ml of the solution, which was gavaged at a dose of 1 cc after each exercise session. The available strains were ready to use and were stored in the refrigerator under cold chain conditions. To prevent temperature shock during gavage, they were kept at room temperature for half an hour before the start of the exercise.

Sacrifice

Forty-eight hours after the last exercise session, the animals were transferred to Kimia Andisheh Teb Biotechnology and Genetics Company, registered under number 205932, for necessary tests and sampling. The euthanasia procedures, including blood collection and tissue sampling (liver), were conducted within 8 hours by a veterinarian and the accompanying team. Blood samples were collected in two forms: whole blood and citrate, for biochemical, enzymatic, and oxidative tests. Liver tissue samples were also collected for the analysis of IL-15 gene expression using the Real-Time PCR method.

Laboratory measurements

For the measurement of glucose and insulin index, serum samples were centrifuged for 15 minutes at a speed of 2500 revolutions per minute. Blood glucose levels were measured using a specific glucose kit (manufactured by Delta Tarh Part with code PI019). The coefficient of variation for this kit was 0.9989. Serum insulin levels were measured using the ELISA laboratory method and a specific kit from the knowledge-based company Kimia Andisheh Teb (STAT FAX 2100, Tehran, Iran).

Insulin resistance was assessed using the Homeostasis Model Assessment (IR-HOMA), which is based on fasting blood glucose (mg/dL) divided by fasting insulin concentration (mU/L) and then divided by a constant of 405. The activity level of the enzyme GPX was measured using an ELISA kit (OD-Multiple, CUSABIO, Houston, USA) according to the protocol provided by the purchasing company.

Expression of IL-15

To investigate the expression of IL-15 genes in each group, the Real-Time PCR technique was used. For primer preparation, 10 microliters of lyophilized primer in distilled water, 0.5 microliters of forward primer, 0.5 microliters of reverse primer, 1 microliter of cDNA, and 8 microliters of diethyl pyrocarbonate (DEPC) water were used. For gene expression analysis, total RNA was extracted using the Qiazol solution based on the protocol provided by SinaGen. The quality of the extracted RNAs was assessed using a spectrophotometer. To prepare single-stranded cDNA, Oligo dt primers and reverse transcription enzyme were used according to the relevant protocol. Each PCR reaction was performed in the ABI Step One device following the manufacturer's protocol. The reaction cycles for Real-Time PCR for the IL-15 gene were conducted at three temperatures: 94°C, 60°C, and 72°C. A melting curve analysis was performed to verify the accuracy of the PCR reactions. Glyceraldehyde 3-phosphate dehydrogenase (GAPDH) was used as the reference gene for IL-15. The expression levels of control and experimental genes were measured simultaneously. The primers used are listed in Table 1.

Table 1. Characteristics of the primers used in the study

Gene	Primer sequence
IL-15	F: 5 GTAGGTCTCCCTAAAACAGAGGC 3 R: 5 TCCAGGAGAAAGCAGTTCATTGC 3
GAPDH	F: 5 GTAGGTCTCCCTAAAACAGAGGC 3 R: 5 TCCAGGAGAAAGCAGTTCATTGC 3

Statistical analysis

Data were presented as mean ± standard deviation. The normality of the data was confirmed using the Shapiro-Wilk statistical test. To examine the variations of the research variables among different groups, a one-way ANOVA statistical test was used. Additionally, the Tukey post hoc test was employed to investigate the changes of each variable between the groups. Pearson correlation was used to assess the correlation between serum GPX and hepatic IL-15. Data were analyzed using SPSS statistical software version 26, and GraphPad Prism 9 was used for plotting the graphs. A significance level of $p < 0.05$ was considered.

Results

Changes in the glycemic indices of animals in different research groups are shown in Figure 1. The results of the one-way ANOVA statistical test indicated that there is a significant difference in serum glucose levels among the various research groups ($F=6.080, p=0.0009$). The results of the Tukey post-hoc test showed that the HFD, HFD+Tr, and HFD+Sup groups had a significant increase in serum glucose compared to the healthy control group ($p<0.05$). However, the changes in the combined treatment group (HFD+Tr+Sup) were not significant compared to the control and HFD groups (Figure 1A).

The results of the one-way ANOVA statistical test indicated that there is a significant difference in serum insulin levels among the various research groups ($F=3.005, p=0.0327$). The results of the Tukey post-hoc test showed that the HFD+Tr+Sup group had a

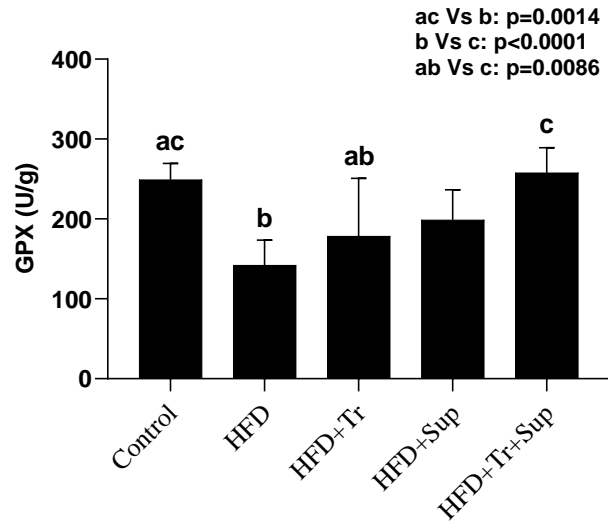


Figure 2. Changes in the antioxidant index GPX in different research groups. Data are presented as mean ± standard deviation. A significance level of $p < 0.05$ is considered. Abbreviations: HFD: high-fat diet, Tr: exercise, Sup: supplement.

significant decrease in serum insulin compared to the HFD+Tr group ($p<0.05$) (Figure 1B). The results of the one-way ANOVA statistical test indicated that there is a significant difference in HOMA-IR values among the various research groups ($F=3.212, p=0.0252$). The results of the Tukey post-hoc test showed that the HFD+Tr group had a significant increase in HOMA-IR compared to the healthy control group ($p<0.05$) (Figure 1C).

Changes in the antioxidant index GPX in different research groups are shown in Figure 2. The results of the one-way ANOVA statistical test indicated that there is a significant difference in the GPX antioxidant index among the different research groups ($F=8.942, p<0.0001$). The results of the Tukey post-hoc test showed that the HFD groups had a significant decrease in GPX compared to the healthy control group ($p<0.05$). However, only

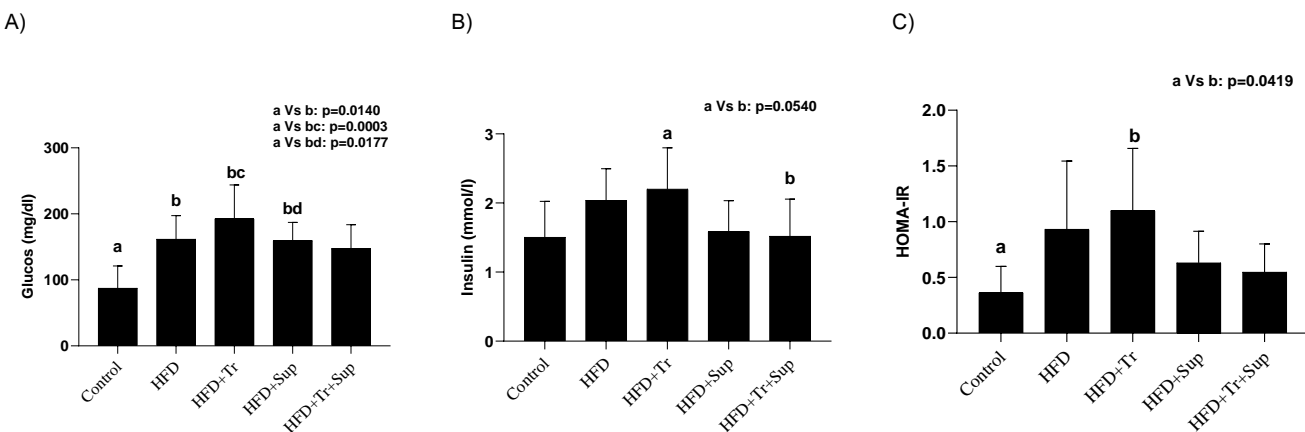


Figure 1. Changes in glucose levels (A), insulin (B), and HOMA-IR © in different research groups. Data are presented as mean ± standard deviation. A significance level of $p < 0.05$ is considered. Abbreviations: HFD: high-fat diet, Tr: exercise, Sup: supplement.

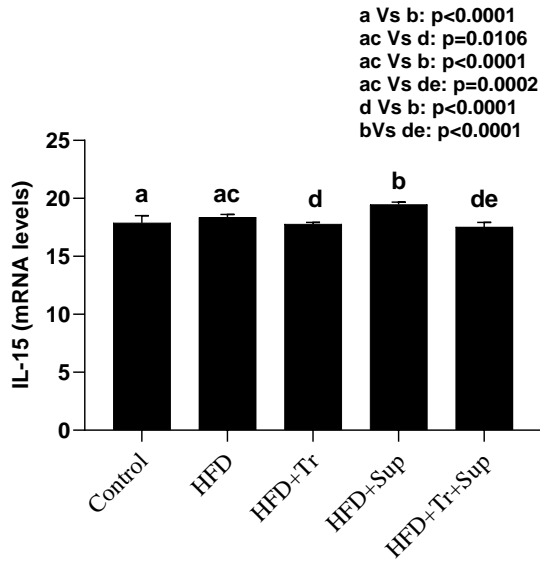


Figure 3. Changes in IL-15 gene expression in different research groups. Data are presented as mean ± standard deviation. A significance level of $p < 0.05$ is considered. Abbreviations: HFD: High-Fat Diet, Tr: Training, Sup: Supplement.

the HFD+Tr+Sup group showed a significant increase in GPX compared to the HFD and HFD+Tr groups ($p < 0.05$).

Changes in IL-15 gene expression in different research groups are shown in Figure 3. The results of the one-way ANOVA statistical test indicated that there is a significant difference in IL-15 gene expression among the various research groups ($F=42.56, p < 0.0001$). Based on the Tukey post hoc test, the HFD

+Sup group showed a significant increase in IL-15 gene expression compared to the healthy control group. Compared to the HFD group, the HFD+Tr and HFD+Tr+Sup groups showed a decrease, while the HFD+Sup group exhibited a significant increase in IL-15 gene expression ($p < 0.05$). Additionally, the HFD+Tr+Sup group demonstrated a significant decrease in this gene compared to the HFD+Sup group ($p < 0.05$).

To examine the correlation between the antioxidant index GPX and the expression of the IL-15 gene, Pearson correlation test was used. The results of this test indicated that there was no correlation between GPX and IL-15 gene expression in the different research groups (Figure 4).

Discussion

Interleukin-15 (IL-15) is expressed in many tissues throughout the body. This factor exhibits different functions in various tissues as well as in physiological and pathological conditions. Generally, it has been stated that IL-15 inhibits apoptosis and promotes cell proliferation and differentiation. However, the role of IL-15 in the liver remains unknown. Therefore, our objective was to determine whether IL-15, under pathological conditions induced by a high-fat diet, contributes to the expansion of inflammation and the weakening of the antioxidant system. Additionally, we aimed to investigate whether exercise and probiotics could effectively regulate this gene, enhance the antioxidant system, and influence glycemic indices.

High-fat diets lead to an increase in circulating free fatty acids, which can elevate ROS levels. This overproduction of ROS results in oxidative damage to macromolecules such as DNA, pr-

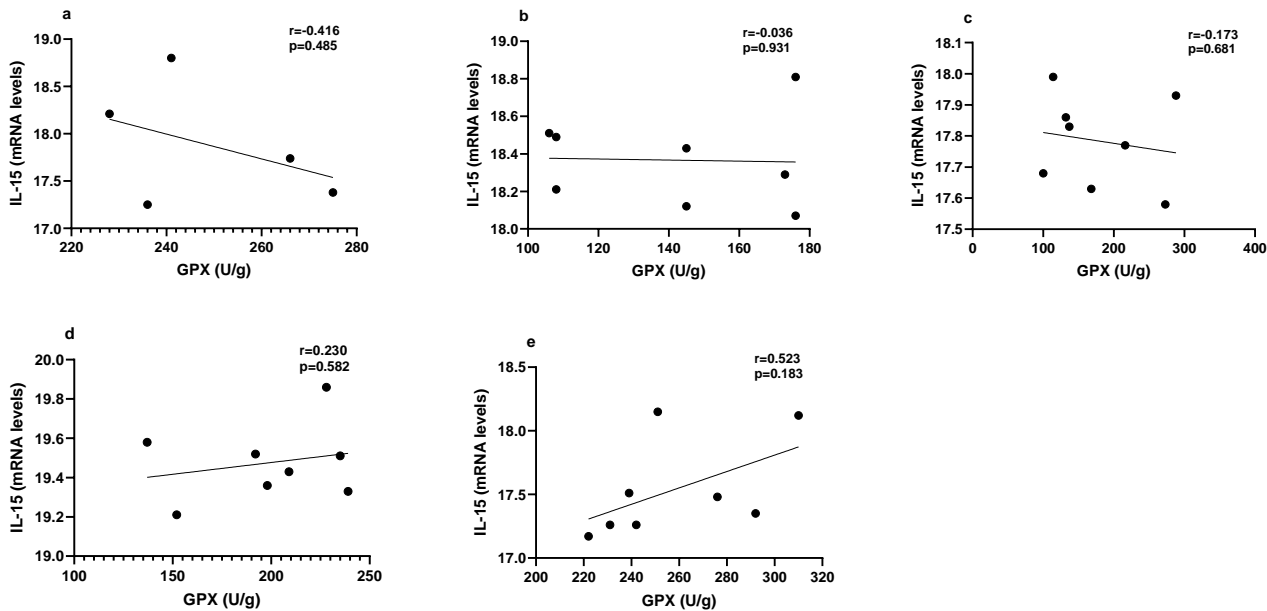


Figure 1. Correlation between antioxidant index GPX and IL-15 gene expression in different research groups. a: Control group, b: HFD group, c: HFD+Tr group, d: HFD+Sup group, e: HFD+Tr+Sup group. Data are presented as mean ± standard deviation. A significance level of $p < 0.05$ is considered. Abbreviations: HFD: high-fat diet, Tr: exercise, Sup: supplement.

-oteins, and lipids, contributing to cellular dysfunction and inflammation (Tan & Norhaizan, 2019). Consumption of a high-fat diet is associated with increased lipid peroxidation, evidenced by elevated levels of malondialdehyde (MDA) and other oxidative stress markers (GPx). This process damages cell membranes and promotes further oxidative stress (Jiang et al., 2021). The high levels of free fatty acids and ROS stimulate immune cells, leading to the secretion of pro-inflammatory cytokines. This systemic inflammation can initiate a chronic low-grade inflammatory state, which is linked to various metabolic disorders (Tan & Norhaizan, 2019). Therefore, the increase in inflammatory factors and the decrease in antioxidant capacity in the HFD group seem reasonable. Therefore, exercise and probiotic supplementation can be effective in controlling inflammation and oxidation of HFD.

The results of the present study indicated that the combination of exercise and probiotic supplementation played a significant role in insulin regulation. Although it did not lead to a significant reduction in blood glucose levels compared to the HFD group, it brought blood glucose levels closer to those of the healthy control group. Additionally, the combination of exercise and probiotics increased GPX levels while decreasing IL-15 levels in liver tissue. Aerobic exercise is known to enhance antioxidant defenses, potentially increasing GPx activity (Ye et al., 2021), which helps reduce oxidative damage by reducing hydrogen peroxide levels. Probiotics may further modulate inflammatory responses (Cristofori et al., 2021), possibly through the gut microbiome, which can influence systemic inflammation and immune responses, including IL-15 production. The gut microbiome is a diverse community of bacteria that carry out various functions influencing overall host health, including nutrient metabolism, immune system regulation, and defense against infection. Certain gut bacteria are associated with inflammatory molecules that may trigger inflammation in various body tissues. Inflammation underlies many chronic conditions like obesity, atherosclerosis, type 2 diabetes, and inflammatory bowel disease (Al Bander et al., 2020). Inflammation can be triggered by structural components of bacteria, resulting in a cascade of inflammatory pathways involving interleukins and other cytokines. Metabolic by-products of bacteria, including some short-chain fatty acids, can also play a role in inhibiting inflammatory processes and controlling IL-15 (Al Bander et al., 2020). Suzuki et al. (2006) demonstrated that IL-15 expression increases during liver injury, and IL-15 treatment induces a wound healing response in healthy adult mice. These findings suggest that IL-15 may assist in the remodeling activity in damaged liver tissue. In the current study, a high-fat diet model was used, resulting in increased IL-15 levels. Therefore, this increase is not solely due to remodeling effects but may also stem from the inflammatory role of this factor. In the bloodstream, GPX levels were also reduced in the HFD group. Although the correla-

-tion between IL-15 and GPX was not confirmed, it seems that the inflammation induced by HFD may contribute to the weakness of the antioxidant system, as glucose levels were also elevated in this group, indicating that this increase plays a role in the expansion of inflammation and the weakness of antioxidant enzymes such as GPX. In contrast, exercise and probiotics had a significant role in regulating GPX and IL-15 levels in liver tissue. Contrary to the findings of the present study, Ziloubaf et al. (2022) showed in their study that probiotic consumption and moderate-intensity continuous training significantly increased IL-15 levels in a model of non-alcoholic steatosis (Ziloubaf et al., 2022). In the current study, HFD was used to induce liver disease, whereas Ziloubaf and colleagues used tetracycline for model induction, and the type of exercise in the two studies was different; thus, the differences in results seem reasonable. Previous studies suggest that both endurance and resistance exercise can raise IL-15, but the effects may differ based on the type of exercise performed. Exercise intensity and duration play crucial roles in influencing interleukin-15 (IL-15) levels, which are important for metabolic and immune functions. Higher intensity exercises, such as eccentric resistance training, have been shown to elicit greater increases in IL-15 compared to lower intensity activities. For instance, studies indicate that eccentric resistance exercise can lead to significant elevations in IL-15 levels, particularly in athletes (Vasconcelos et al., 2018). However, we use an aerobic exercise for the treated rats. The increase in IL-15 is transient, with levels peaking shortly after exercise and returning to baseline within a few hours. Typically, IL-15 levels can remain elevated for up to 120 minutes post-exercise, but they generally revert to pre-exercise levels within three hours (Khalafi et al., 2024). While acute exercise significantly boosts IL-15 levels, chronic exercise training has shown inconsistent effects on circulating IL-15. Some studies report no significant changes in IL-15 levels following long-term training, suggesting that the acute response is more pronounced than any chronic adaptations (Khalafi et al., 2024). Therefore, exercise intensity and duration are critical factors influencing IL-15 levels. Acute, high-intensity exercises lead to significant and immediate increases in IL-15, while chronic training has variable effects. The transient nature of the IL-15 response underscores its role as a myokine involved in metabolic and immune regulation during and after exercise.

In the present study, considering the increase in GPX and the decrease in IL-15 in the exercise and probiotic treatment group, particularly in the combined treatment, it appears that the role of exercise and probiotics in controlling inflammation and enhancing the antioxidant system has been very effective.

Different probiotic strains have been shown to influence glutathione peroxidase (GPx) levels, which is an important antioxidant enzyme involved in protecting cells from oxidative stress. *Lactobacillus plantarum* has been reported to enhance the expression of GPx in various studies. It is known for its antiox-

-idant properties, which help in mitigating oxidative stress by increasing the activity of antioxidant enzymes, including GPx (Averina et al., 2021). Various probiotic strains, particularly Lactobacillus and Bifidobacterium species, can positively influence glutathione peroxidase levels and enhance the antioxidant defense system. The specific effects can vary depending on the strain and the context of use, such as the type of oxidative stress encountered. Probiotics like Lactobacillus plantarum and Lactococcus lactis MG5125 have shown promising results in increasing GPx activity, while other strains may contribute to overall oxidative stress reduction through different mechanisms. Consistent with these results, Wang et al. (2016) confirmed in their study that the probiotic L. casei Zhang reduces pro-inflammatory cytokine production and hepatic inflammation induced by LPS/GalN through modulation of the TLR-MAPK-PPAR- γ signaling pathways and gut microbiota (Wang et al., 2016). Several important mechanisms underpin the beneficial effects of probiotics, including the modification of gut microbiota, competitive adhesion to the mucosa and epithelium, strengthening the epithelial gut barrier, and regulating the immune system and inflammation (Bermudez-Brito et al., 2012; Fontana et al., 2013). Most of these mechanisms involve the regulation of gene expression in specific tissues, particularly the gut and liver. Exercise can also amplify these anti-inflammatory effects. Regular exercise can be considered an effective strategy for treating liver diseases. In non-alcoholic fatty liver disease, exercise plays a significant role by reducing hepatic fat content, increasing beta-oxidation of fatty acids, inducing protective autophagy in the liver, upregulating PPAR- γ expression, as well as decreasing hepatocyte apoptosis and increasing insulin sensitivity (Farzanegi et al., 2019). Exercise also suppresses excessive ROS and oxidative stress in NAFLD by enhancing the regulation of several antioxidant enzymes and anti-inflammatory mediators (Farzanegi et al., 2019). Given these results, exercise and probiotics play an important role in liver diseases induced by a high-fat diet, and more detailed studies are needed to explore the relationship between the inflammatory and antioxidant systems in the liver.

Conclusion

It seems that exercise training, especially aerobic, may regulate both circulating IL-15 and GPx levels separately. Probiotics that enhance GPx activity in the liver may also influence IL-15 levels, either directly or indirectly, through their effects on oxidative stress and inflammation. The combination of exercise and probiotic nutrition may synergistically modulate both IL-15 in the liver, leading to improved metabolic health outcomes. In addition, its better the GPx activity were measure in the level and then consider the correlation between them with theses treatments. Based on these results it seems that combination of probiotics and aerobic exercise can lead to greater improvements in weight,

glucose tolerance, and fat composition compared to either intervention alone. This suggests a synergistic effect that enhances the overall benefits of both in reducing side effects of obesity.

What is already known on this subject?

Probiotics are currently used for the treatment or prevention of human diseases, conditions, and syndromes. They have also been shown to have a positive impact on neuroinflammation and pain, as well as on infections from seasonal diseases.

What this study adds?

Exercise training, may regulate both circulating IL-15 and GPx levels separately. Probiotics that enhance GPx activity in the liver may also influence IL-15 levels, either directly or indirectly, through their effects on oxidative stress and inflammation.

Organ Cross-Talk Tips:

- Aerobic exercise and probiotic supplementation can improve crosstalk between serum antioxidant and liver anti-inflammatory factors.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval This study was conducted under the supervision of the Animal Ethics Committee of the Islamic Azad University, Science and Research Branch, Tehran, Iran, with ethical code IR.IAU.SRB.REC.1402.188.

Informed consent Not applicable

Author contributions

Conceptualization: M.P, M.GH, H.A, F.GH.; Methodology: M.P, M.GH.; Software: H.A, F.GH.; Validation: M.GH.; Formal analysis: H.A, F.GH.; Investigation: M.GH.; Resources: M.P.; Data curation: M.P, M.GH, H.A, F.GH.; Writing - original draft: M.P.; Writing - review & editing: M.P, M.GH, H.A, F.GH.; Visualization: M.P, M.GH, H.A, F.GH.; Supervision: M.GH.; Project administration: M.GH.; Funding acquisition: M.GH.

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