Medical Laboratory Journal
Online ISSN: 2538-4449
Golestan University of Medical Sciences

Original Article

Response of Cardiac Tissue β-catenin and GSK-3β to Aerobic Training and Hyaluronic Acid in Knee OA Model Rats

Hadi Alinezhad
Department of Exercise Physiology Ayatollah Amoli Branch, Islamic Azad University, Amol, Iran
Asieh Abbassi-Daloii
Department of Exercise Physiology Ayatollah Amoli Branch, Islamic Azad University, Amol, Iran
Parvin Farzanegi
Department of Exercise Physiology Sari Branch, Islamic Azad University, Sari, Iran
Ahmad Abdi
Department of Exercise Physiology Ayatollah Amoli Branch, Islamic Azad University, Amol, Iran
Corresponding author: Asieh Abbassi-Daloii
Email: abbasi.daloii@gmail.com
Tel: +98 09111274366
Address: Department of Sport Physiology, Ayatollah Amoli Branch, Islamic Azad University, Amol, Iran

Received: 2019/10/30
Revised: 2020/01/20
Accepted: 2020/01/20

DOI: 10.29252/mlj.15.1.45

ABSTRACT

Background and objective: *Osteoarthritis* (*OA*) and *cardiovascular disease* (*CVD*) are highly prevalent. The purpose of the present study was to investigate the effect of regular aerobic training and hyaluronic acid on cardiac tissue Wnt signaling pathway in experimental model of knee *OA*.

Methods: 42 male rats were divided into 6 groups (7 in each group): 1) control, 2) patient, 3) salin, 4) HA, 5) exercise, and 6) exercise + HA. In the training groups, the *OA* model was first induced, followed by 5 days of running on the treadmill for 5 weeks. Hyaluronic acid was injected intra-articularly. After 12 to 14 hours of fasting and 72 hours after the last training session, cardiac tissue sampling was performed for β-catenin and glycogen synthase kinase-3 (GSK-3β) analysis. The expression of the β-catenin and GSK-3β genes in the cardiac tissue was analyzed by RT-PCR. Data analysis was performed using one-way ANOVA if a significant difference was observed by Tukey's post hoc test ($P < 0.05$).

Results: Induction of *OA* in rats led to a significant increase in β-catenin gene and a significant decrease in cardiac tissue GSK3 gene compared to healthy control group. The results also showed that regular aerobic training, hyaluronic acid injection, and a combination of both treatments reduced the cateninβ gene and increased the cardiac tissue GSK3 gene compared to the rats of *OA* group.

Conclusion: Regular aerobic training in combination with hyaluronic acid may exert its protective effect by reducing the expression of β-catenin and increasing the expression of cardiac tissue GSK-3β gene; this may be caused by the heart disease in the model, empirically preventing osteoarthritis.

Key words: *Osteoarthritis,* Cardiovascular disease, Exercise, Hyaluronic acid, beta catenin

INTRODUCTION

OA is an advanced degenerative process that causes the irreversible loss of articular cartilage and is characterized by pain and dysfunction of the joint. It is the most common arthritic disease and one of the most prevalent chronic diseases (1). *CVD* is also the most common cause of death in developed countries (2). *OA* and *CVD* increase with age, so they are commonly co-occurring diseases(1). There are studies showing a strong independent relationship between *OA* and *CVD* (3). Recently, new meta-analyses have shown that *OA* is an important risk factor for *CVD* (4).

Wnt/β-catenin, which is inactive in many adult organs, is activated in response to injury (5). Its role in the recovery and regeneration of complex and incomplete tissue is understood although a growing number of data indicate that its activation results in increased fibrotic healing (6). Recent studies have shown that the short-term inhibition of Wnt therapy after ischemic heart injury improves the regeneration associated with less fibrosis (7). In the pathogenesis of *OA*, activation of focal Wnt signaling was observed in both the synovial and cartilage after injury with the increase in the expression of Wnt ligands and target genes (8).

The focal Wnt signaling pathway is involved in the β-catenin multifunctional protein. β-catenin is known for its function in cell adhesion through interaction with the cadherin family membrane transport proteins, but here, it acts as a signaling mediator. In the absence of Wnt, the cytosolic β-catenin interacts with a degraded complex consisting of Axin, APC, and GSK3β, thereby phosphorylated and subsequently degraded by the proteasome. In the presence of a Wnt ligand, the degradation complex is cleaved and β-catenin is stabilized, enters the nucleus, and regulates the transcription of target genes through interaction with TCF/LEF transcription factors (9).

Today, physical activity is widely recommended by health care professionals for the prevention and management of chronic health conditions, including cardiovascular diseases, mental illnesses, and obesity (10). However, the effect of walking on the development or progression of osteoarthritis is inconsistent. On the other hand, some studies have shown that walking has protective effects on the loss of articular space. In contrast, older adults who performed high levels of heavy physical activity had an increased risk of radiographic knee *OA* (10).

A meta-analysis of 33 studies found that people who did 150 minutes of weekly moderate-intensity physical activity had a lower risk (14%) of developing *CVD* similar to those who did 300 minutes of moderate-intensity exercise per week. Moderate-to-moderate physical activity is 20% less likely to lead to *CVD* (11). Even light-to-moderate activity (at least one hour of walking per week) is associated with lower rates of *CVD* (12, 13).

There are very few studies regarding the effects of aerobic and endurance training on Wnt signaling pathway and its proteins such as β-catenin and GSK3β, for which conflicting results have often been reported. Fuji Maki et al. observed that 4 weeks of voluntary, slow rotation cycling induced the over-regulation of the Wnt pathway and increased β-catenin in the exercise group; this caused the activation of satellite cells and increased the transcription of myogenic genes in the training group (14, 15, 16, 17). However, Amin et al. found no change in the expression of β-catenin protein following exercise in rats (18).

Hyaluronic acid, as a polymer, comprises a major part of the extra-cellular matrix in the connective tissue. This polysaccharide is an important extracellular matrix glycosaminoglycan with myriad properties, such as binding to water molecules, stores for growth factors, receptor binding, angiogenesis, proliferation, cell migration, and repair of damaged areas (19). Intra-articular injection of hyaluronic acid is one of the most common treatments for osteoarthritis, particularly in patients who have failed other treatments due to toxicity or ineffectiveness. Many studies have shown the beneficial effects associated with the intra-articular injection of hyaluronic acid on pain management and patient activity (19).

Given the advantageous effects of aerobic training and hyaluronic acid, this study aimed

to compare the responses of cardiac tissue β-catenin and GSK-3β to aerobic training and hyaluronic acid in knee *OA* model rats.

MATERIALS AND METHODS

In the present experimental study, the subjects (mice) were in control of many variables in the laboratory.

Ethical code: NO.19.33.2018

The statistical samples were 42 male Wistar rats with a mean weight of 300-250 g. These animals were divided into 6 groups (7 in each group): 1) control, 2) patient, 3) salin, 4) HA, 5) exercise, and 6) HA + exercise.

After transferring the animals to the laboratory, the subjects were housed in transparent polycarbonate cages measuring 15*15*30 cm (Razi Rad Company) with an ambient temperature of 20±2° C, 50±5% humidity, and proper ventilation. The feed was made from pellet obtained from Karaj Animal Feed Company and was supplied ad libitum in a 500 ml bottle for laboratory animals.

***OA*:** In order to induce the osteoarthritis model, mice were first anesthetized with ketamine and xylazine, and a vertical incision was then made in the inner part of the right knee on the pre-shaved skin. After removing the skin, the inner lateral ligament of the knee was removed to allow for internal meniscus; next, incision was performed, incompletely resulting in rupture and injury to the meniscus. The *OA* model was induced and again sutured by sterile method. All procedures were performed in the least amount of pain to the animal and in accordance with the principles of working with laboratory animals. After induction, a recovery period of three weeks was considered. During this period, the rats were operated on the treadmill for 4 daysfor 5 to 10 minutes at a speed of 6 to 8 m/min and a slope of zero percent. The training program consisted of 25-29 minutes of running on a treadmill with no slope at 15 m/min for the first week, progressively overloading the following weeks for a period of 34-44 minutes. The intensity reached 16-18 m/min in the fourth week. Five minutes before and after training were also allocated to warming up and cooling down the animals. In the hyaluronic acid receiving groups, two successive intra-articular injections were administeredwith a one-week interval. Intra-articular injections of 25 µL of HA (Hyalgan® sodium hyaluronate; Fidia Farmaceutici S.p.A., AbanoTerme, Italy) was administered using a 27 gauge, 0.5 inch needle (20).

After performing the research to eliminate the acute effects of exercise, all animals were sampled in completely similar conditions, followed by 12 to 14 hours of fasting and 72 hours after the last training session. Mice were anesthetized by the intraperitoneal injection of a combination of ketamine (70 mg/kg) and xylazine (3–5 mg/kg). According to the predetermined timing, cardiac tissue sampling was done on the control and exercise groups, which were washed with saline in special tubes and then frozen in liquid nitrogen.

The expression of the β-catenin and glycogen synthase kinase-3 genes in the cardiac tissue was analyzed by RT-PCR. First, the tissue samples were homogenized in phosphate buffer (pH 7.0) at 4 °C with a homogenizer. Total RNA was extracted from the samples using the RNX-Plus kit (SinaClon; RN7713C). Nanodrop ND-1000 spectrophotometer (Thermo Sci., Newington, NH) was used to estimate the quantity and quality of the extracted RNA. Synthesis of cDNA was done using the Revert Aid Reverse Transcriptase (Thermo science, Germany) at 42 °C for 1 hour (Thermo science, Germany). For amplifications, a Rotor Gene 6000 (Corbett Research, Australia) thermocycler and Real Q-PCR 29 Master Mix Kit (Amplicon, Denmark) in 40 cycles were applied. Primer sequences were as follows: beta-catenin, forward: 5′-ATGCTGAGGAAGAAGATGTGGA-3′, reverse: 5′-ATGAAACTGCGTGGATGGGA-3′;glycogen synthase kinase-3, forward: 5′-CAAAGCAGCTGGTCCGAGG-3′, reverse:5′-TCCACCAACTGATCCACACCAC-3′ ; glyceraldehyde 3-phosphate dehydrogenase (GAPDH),forward: 5′-GGATAGTGAGAGCAAGAGAGAGG-3′,reverse:5′-ATGGTATTGGAGAGAAGGGAGGG -3′. The level of mRNA was normalized to the amount of GAPDH mRNA.

Descriptive statistics were used to classify the data. Shapirovilk test was used to specify the normality of the data distribution. One-way analysis of variance determined the significance of the differences between the variables and their interaction. Tukey post hoc

test was used to determine the significance of the data. The results were analyzed at 95% confidence level (P≤0.05) and IBM SPSS statistics version 20 was employed for statistical analysis. All data were presented as mean±SD.

RESULTS

GSK3: Results of ANOVA test for the expression of heart tissue GSK3 in the study groups and the calculated F value (5.489) and its significance at P = 0.000 level indicated a significant difference in the expression of GSK3 heart tissue among different study groups. Based on these findings, the mean expression of GSK3 gene on cardiac tissue in *OA* mice was significantly reduced compared to the controls. Meanwhile, there was an increase in the expression of heart tissue GSK3 gene in the exercise, HA, and exercise + HA groups, compared to the patient group; this increase was especially significant in the HA + exercise group (P = 0.020). There was also a significant difference between HA and exercise + HA groups (P = 0.017)(Table 1 and Figure 1).

β-Catenin: The results of ANOVA test for β-Catenin expression of cardiac tissue in different research groups and the calculated F value (5.531) and its significance at P = 0.000 showed a significant difference in cardiac tissue β-Catenin gene expression among the study groups. Based on Tukey's post hoc test, there was a significant increase in the expression of cardiac tissue β-Catenin gene in *OA* rats compared to the control group. However, in the exercise, HA, and exercise + HA groups, there was a reduction compared to the patients; this decrease was significant in the HA + exercise group (P = 0.042). There was also a significant difference between HA and exercise + HA groups (P = 0.032), (Table 2 and Figure 2).

Table 1: Mean and standard deviation of relative expression of GSK

Groups	mean ± Standard deviation
Control	0.109837678±.0701679164
Patient	0.002812940±.0050248646
Salin	0.001391520±0.0009376190
HA	0.001656296±0.0022045680
Exercise training	0.038627606±0.0273052638
HA + Exercise training	0.078940273±0.0522345671

Figure 1. Relative expression of GSK for research groups

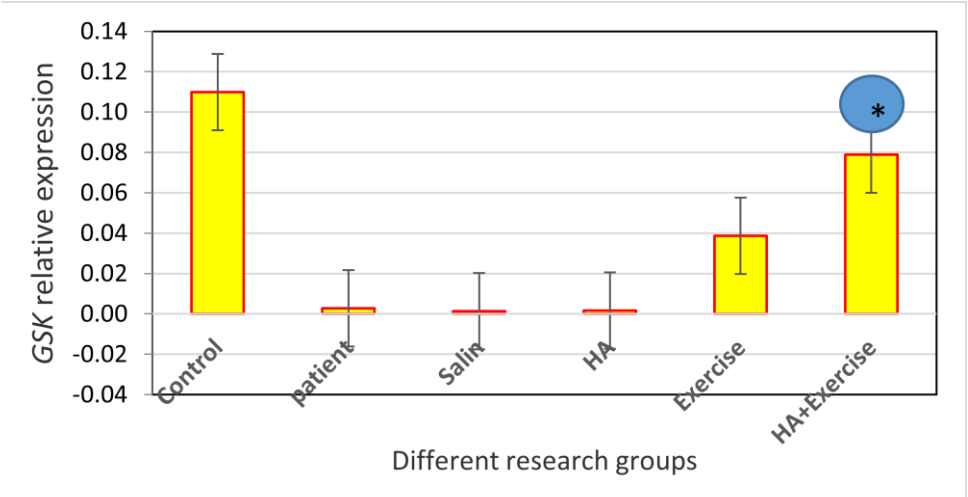
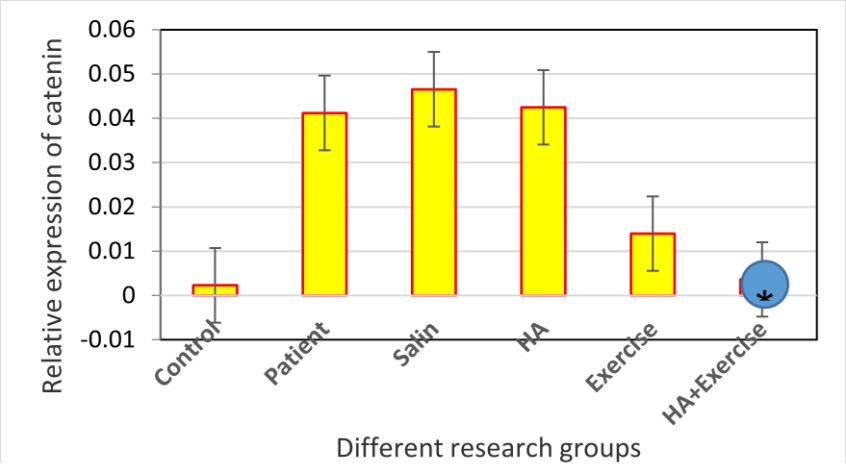


Table 2: Mean and standard deviation of relative expression of β-catenin

Groups	mean ± Standard deviation
Control	0.002294344±0.0012000633
Patient	0.041181219±0.0108733745
Salin	0.046539896±0.0255876530
HA	0.042465583±0.0336801149
Exercise training	0.013968579±0.0094115355
HA + Exercise training	0.003632002±0.0030377625

Figure 2. Relative expression of *catenin* in the study groups
*: Significant sign relative to the patient group &: Significant sign relative to the HA+Exercise group



DISCUSSION

Induction of *OA* in rats led to a significant catenin gene as well as a -increase in β significant decrease in cardiac tissue GSK3 gene compared to the healthy controls, -indicating that the activation of the Wnt/ β catenin is in the heart. Previous studies have shown that activation of the Wnt signaling pathway is common between *OA* and cardiovascular diseases (21). In a study by Lietman et al., Wnt signaling inhibition improved *OA* in the experimental model (22). Yue Zhao et al. reported that Wnt signaling played a key role in hypertension, cardiac hypertrophy, and cardiac fibrosis (23). In Zhao's study, it was also shown that the induction of osteoarthritis activated the Wnt signaling pathway through enhancing β-catenin and depletion of GSK3 in heart tissue, possibly increasing the risk of *CVD* in experimental models of knee *OA*. The results also showed that regular aerobic training, hyaluronic acid injection, and a combination catenin gene -of both treatments reduced the β and increased the cardiac tissue GSK3 gene compared to the patient group. These changes were significant in the combination treatment group.

The results of Habibi et al. showed that the expression level of β-catenin gene in the aerobic training group was insignificant and approximately 30% lower than the control group (24). In agreement with the results of the present study, Amin et al. reported that there was no significant change in the expression of β-catenin protein following exercise in rats (18). Vissing et al. reported that 10 weeks of exercise training did not significantly change the baseline levels of messenger proteins such as Akt-mTORC1, β-catenin, and glycogen synthase kinase-3β (25). Habibi et al. also found that the expression level of glycogen synthase kinase-3β was significantly higher in the aerobic exercise group than in the control group (24).

These findings are inconsistent with the results of the present study, which may be attributed to the differences in the type of subjects and

the tested tissues; they examined Wnt signaling in healthy subjects while the present study evaluated cardiac tissue Wnt signaling in an experimental model of osteoarthritis. Also, differences in duration, intensity, and type of exercise can contribute to these inconsistencies. In the current study, through increasing GSK-3 and reducing beta-catenin aerobic training, depletion GSK3 led to the inactivation of the Wnt signaling pathway, which may prevent heart disease.

Several studies have suggested that GSK-3β acts as a negative regulator of hypertrophy in C2C12 muscle cells (26). However, decreased expression of GSK-3β and its phosphorylation are associated with β-catenin nuclear accumulation. β-catenin nuclear accumulation results in the increased formation of complexes with the LEF and TCF transcription factor families (27). These processes are related to the activation of the transcription of target genes to stimulate the synthesis and repair and regenerate the muscle cells (17, 27). However, in addition to reducing the accumulation of β-catenin, the GSK-3β protein plays a complex role via different messenger proteins in the cell.

In this regard, Fuji Maki et al. proposed that vigorous endurance activities such as intense and prolonged dosing may increase the risk of muscle atrophy and sarcopenia, particularly in the elderly (17). Shin Fujimaki et al. reported that chronic running activated the satellite cells in the diabetic mice, improving the Wnt signaling pathway (28). Pourrazi et al. showed that by affecting Wnt signaling, exercise activity improved the process of muscle mass loss due to dietary restriction (29). In this regard, some reports have suggested that GSK-3β can either directly activate the phosphorylation of pre-apoptotic proteins such as Bax or increase the expression of Bim (30, 31). In other words, by stimulating apoptosis, GSK-3β can pave the way for accelerated muscle atrophy and sarcopenia. Sadeghipour et al. found that 6 weeks of endurance training significantly decreased GSK-3β gene expression in rats with diabetes neuropathy probably due to adaptation to endurance training (32). Based on the results of different studies, a significant relationship exists between exercise and the changes in GSK-3β expression in skeletal muscle and cardiac muscle of diabetic rats. Some studies have shown an exercise-induced decrease in this kinase in the rats' twin muscles; they have also suggested that this decrease is likely caused by the increase in the activity of Akt / mTOR signaling pathways, which are GSK-3β phosphorylation and inactivation pathways (32).

In a study by Young Kim et al, treadmill training increased Wnt3 expression and suppressed GSK-3 expression in diabetic rats. This indicates that treadmill training activates the Wnt signaling pathway, inhibiting GSK-3β expression in diabetic mice. In a Wnt3 activity-dependent behavior, GSK-3 is released from the hippocampal neurons and control hippocampal neurogenesis (33).

Lo et al. observed that intra-articular injection of hyaluronic acid had no effect compared to placebo, which is inconsistent with our study (34). Some studies have suggested that hyaluronic acid not only restores the fluid viscosity but also affects the pathologic factors associated with *OA* (35). Numerous animal studies corroborate the effect of hyaluronic acid on reducing joint cartilage destruction and improving tissue repair (36). In a study by Listrat et al. articular cartilage destruction was lower in patients with *OA* compared with hyaluronic acid injection (37). Nowadays, intra-articular injection of hyaluronic acid is widely used in the treatment of *OA* and pain relief (23). Hyaluronic acid exerts its effects via increasing the number of living chondrocytes, inducing thickening and repair on cartilage surface, preventing nitric oxide production in synovial fluid and miniscule fluid, inhibiting canthropocyte apoptosis, and reducing matrix metalloproteinase-3 and interleukin-1β in synovial fluid (38- 40). In the present study, the combination of hyaluronic acid with regular aerobic exercise had a better effect on the inhibition of Wnt signaling in the heart tissues of the subjects; this may be attributable to the same mechanisms and changes in hyaluronic acid in the knee structure and pain reduction in the subjects. However, definitive results require further studies. Wnt signaling plays different roles in healthy and patient subjects. The activation of the pathway in healthy tissues in order to influence exercise activity in the target tissue seems to have beneficial effects. On the other hand, the findings of this study suggested that the activation of the Wnt signaling pathway in

the heart tissue of *OA* mice might indicate the increased risk of heart disease in osteoarthritic subjects.

CONCLUSION

In the present study, induction of the experimental model of *OA* increased the expression of β-catenin gene and decreased the expression of cardiac tissue GSK-3β; activated Wnt signaling increased the risk of *CVD*. Regular exercise training, hyaluronic acid injection, and combination of both therapies reduced the cardiac tissue β-catenin expression, increased GSK-3β gene expression, and inhibited Wnt signaling in the heart, preventing *CVD* in the experimental model of *OA*.

ACKNOWLEDGEMENT

The authors are grateful to all study participants for their cooperation.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

REFERENCES

1. Karaarslan F, Ozkuk K, Seringec Karabulut S, Bekpinar S, Zeki Karagulle, Nergis Erdogan M. *How does spa treatment affect cardiovascular function and vascularMendothelium in patients with generalized osteoarthritis?* A pilot study through plasma asymmetric di-methyl arginine (ADMA) and L-arginine/ADMA ratio. CrossMark, International Journal of Biometeorology. 2018; 62: 833-842. [DOI:10.1007/s00484-017-1484-0] [PubMed] [Google Scholar]

2. Mahmood SS, Levy D, Vasan RS, Wang TJ. *The Framingham Heart Study and the epidemiology of cardiovascular diseases: a historical perspective.* Lancet. 2014; 383(9921): 999-1008. [DOI:10.1016/S0140-6736(13)61752-3] [PubMed] [Google Scholar]

3. Veronese N, Cereda E, Maggi S, Luchini C, Solmi M, Smith T, et al. *Osteoarthritis and mortality: a prospective cohort study and systematic review with meta-analysis.* Semin Arthritis Rheum. 2016; 46(2):160-167. [DOI:10.1016/j.semarthrit.2016.04.002] [PubMed] [Google Scholar]

4. Wang H, Bai J, He B, Hu X, Liu D. *Osteoarthritis and the risk of cardiovascular disease: a meta-analysis of observational studies.* Sci Rep. 2016; 6(1): 39672. [DOI:10.1038/srep39672] [PubMed] [Google Scholar]

5. Xiao L, Zhou D, Tan RJ, Fu H, Zhou L, Hou FF, et al. *Sustained Activation of Wnt/β-Catenin Signaling Drives AKI to CKD Progression.* J Am Soc Nephrol. 2016; 27(6): 1727-40. [DOI:10.1681/ASN.2015040449] [PubMed] [Google Scholar]

6. Tan RJ, Zhou D, Zhou L, Liu Y. *Wnt/β-catenin signaling and kidney fibrosis.* Kidney Int Suppl. 2014; 4(1): 84-90. [DOI:10.1038/kisup.2014.16] [PubMed] [Google Scholar]

7. Bastakoty D, Saraswati S, Joshi P, Atkinson J, Feoktistov I, Liu J, et al. *Temporary, Systemic Inhibition of the WNT/β-Catenin Pathway promotes Regenerative Cardiac Repair following Myocardial Infarct.* Cell Stem Cells Regen Med. 2016; 2(2):10.16966/2472-6990.111. [DOI:10.16966/2472-6990.111] [PubMed] [Google Scholar]

8. Dell'accio F, De Bari C, Eltawil NM, Vanhummelen P, Pitzalis C. *Identification of the molecular response of articular cartilage to injury, by microarray screening: Wnt-16 expression and signaling after injury and in osteoarthritis.* Arthritis Rheum. 2008; 58(5):1410-1421. [DOI:10.1002/art.23444] [PubMed] [Google Scholar]

9. Gessert S, Kühl M. *The multiple phases and faces of wnt signaling during cardiac differentiation and development*. Circ Res. 2010; 107(2): 186-99. [DOI:10.1161/CIRCRESAHA.110.221531] [PubMed] [Google Scholar]

10. Zhu Z, Aitken D, Cicuttini F, Jones G, Ding C. *Ambulatory activity interacts with common risk factors for osteoarthritis to modify increases in MRI-detected osteophytes.* Osteoarthritis Cartilage. 2019; 27(4): 650-658. [DOI:10.1016/j.joca.2018.12.023] [PubMed] [Google Scholar]

11. Nicola's Lillo, Gonzalo Palomo-Ve'lez, Eduardo Fuentes, Iva'n Palomo. *Role of physical activity in cardiovascular disease prevention in older adults.* Sport Sci Health. 2015; 11(3): 1-7. DOI 10.1007/s11332-015-0233-1. [Google Scholar]

12. Sattelmair J, Pertman J, Ding EL, Kohl HW 3rd, Haskell W, Lee IM. *Dose response between physical activity and risk of coronary heart disease: a meta-analysis.* Circulation. 2011; 124: 789-795. [DOI:10.1161/CIRCULATIONAHA.110.010710] [PubMed] [Google Scholar]

13. Artinian NT, Fletcher GF, Mozaffarian D, Kris-Etherton P, Van Horn L, Lichtenstein AH, et al. *Interventions to promote physical activity and dietary lifestyle changes for cardiovascular risk factor reduction in adults: a scientific statement from the American Heart Association.* Circulation. 2010; 122: 406-441 [DOI:10.1161/CIR.0b013e3181e8edf1] [PubMed] [Google Scholar]

14. Spillanea M, Schwarz N, Willoughby DS. *Upper-body resistance exercise augments vastuslateralis androgen receptor-DNA binding andcanonical Wnt/β-catenin signaling compared tolower-body resistance exercise in resistance-trainedmen without an acute increase in serumtestosterone.* Steroids. 2015; 98: 63-71. [DOI:10.1016/j.steroids.2015.02.019] [Google Scholar]

15. Leal ML, Lamas L, Aoki MS, Ugrinowitsch C, Ramos MSC, Tricoli V, et al. *Effect of differentresistance-training regimens on the WNT-signalingpathway*. Eur J Appl Physiol 2011; 111: 2535-45. [DOI:10.1007/s00421-011-1874-7] [Google Scholar]

16. Brack AS, Conboy MJ, Roy S, Lee M, Kuo CJ,Keller C. *Increased Wnt signaling during agingalters muscle stem cell fate and increasesfibrosis.* Science. 2007; 317(5839): 807-10. [DOI:10.1126/science.1144090] [PubMed] [Google Scholar]

17. Fujimaki S, Hidaka R, Asashima M, Takemasa T, Kuwabara1 T. *Wnt-Mediated Satellite Cell Conversion in Adult and Aged MiceFollowing Voluntary Wheel Running. JBC Papers in Press.* Published on January 30. 2014; M113.539247. [PubMed] [Google Scholar]

18. Amin H, Vachris J, Hamilton A, Steuerwald N, Howden R, Arthur ST. *GSK3β inhibition andLEF1 upregulation in skeletal muscle following about of downhill running.* J Physiol Sci. 2014; 64(1): 1-11. [DOI:10.1007/s12576-013-0284-5] [PubMed] [Google Scholar]

19. Hashemi SS, Rajabi S, Mahmoudi R, Ghanbari A, Jafari Barmak M. *The Investigation of Proliferation of Fibroblasts on¿Chitosan Scaffold in the Presence of Hyaluronic Acid.* Armaghane-danesh. 2018; 23(2): 134-145. [PubMed] [Google Scholar]

20. Suhaeb AM, Naveen S, Mansor A, Kamarul T. *Hyaluronic acid with or without bone marrow derived-mesenchymal stem cells improves osteoarthritic knee changes in rat model: a preliminary report*. Indian J Exp Biol. 2012; 50(6): 383-90. [PubMed] [Google Scholar]

21. Fernandes GS, Valdes AM. *Cardiovascular disease and osteoarthritis: common pathways and patient outcomes.* Eur J Clin Invest. 2015; 45(4): 405-14. [DOI:10.1111/eci.12413] [Google Scholar]

22. Lietman C, Wu B, Lechner S, Shinar A, Sehgal M, Rossomacha E, et al. *Inhibition of Wnt/β-catenin signaling ameliorates osteoarthritis in a murine model of experimental osteoarthritis.* JCI Insight. 2018; 3(3): e96308. [DOI:10.1172/jci.insight.96308] [PubMed] [Google Scholar]

23. Zhao Y, Wang C, Wang C, Hong X, Miao J, Liao Y, et al. *An essential role for Wnt/β-catenin signaling in mediating hypertensive heart disease.* Sci Rep. 2018; 8(1): 8996. [DOI:10.1038/s41598-018-27064-2] [PubMed] [Google Scholar]

24. Bashiri J, NourAzar A, Purrazi H. *Effect of three months aerobic training on Wnt-signaling pathway in skeletal muscle of male rats.* RJMS. 2017; 24(160):7-16 [Google Scholar]

25. Vissing K, McGee S, Farup J, KjolhedeT, Vendelbo M, Jessen N. *Differentiated mTOR butnot AMPK signaling after strength vs enduranceexercise in training-accustomed individuals.* Scand J Med Sci Sports. 2013; 23(3): 355-66. [DOI:10.1111/j.1600-0838.2011.01395.x] [Google Scholar]

26. Rommel C, Bodine SC, Clarke BA, Rossman R, Nunez L, Stitt TN, et al. *Mediation of IGF-1-induced skeletal myotube hypertrophy by PI(3)K/Akt/mTOR andPI(3)K/Akt/GSK3 pathways.* Nat Cell Biol. 2001; 3:1009-13. [DOI:10.1038/ncb1101-1009] [PubMed] [Google Scholar]

27. Petropoulos H, Skerjanc IS. *Beta-catenin is essential and sufficient for skeletal myogenesis in P19 cells.* J Biol Chem. 2002; 277: 15393-9. [DOI:10.1074/jbc.M112141200] [PubMed] [Google Scholar]

28. Fujimaki S, Wakabayashi T, Asashima M, Takemasa T, Kuwabara T. *Treadmill running induces satellite cell activation in diabetic mice.* Biochem Biophys Rep. 2016; 8: 6-13. [DOI:10.1016/j.bbrep.2016.07.004] [PubMed] [Google Scholar]

29. Pourrazi H, Asgharpour-arshad M. *Effect of dietary restriction with or without exercise training on β-catenin and glycogen synthase kinase-3β gene expression in skeletal muscle of male rats.* Razi J Med Sci. 2019: 26(1): 78-88. [Google Scholar]

30. Linseman DA, Butts BD, Precht TA, PhelpsRA, Le SS, Laessig TA, et al. *Glycogen synthasekinase-3β phosphorylates Bax and promotes itsmitochondrial localization during neuronalapoptosis.* J Neurosci. 2004; 24: 9993-10002. [DOI:10.1523/JNEUROSCI.2057-04.2004] [PubMed] [Google Scholar]

31. Beurel E, Jope RS. *The paradoxical pro- andanti-apoptotic actions of GSK3 in the intrinsic andextrinsic apoptosis signaling pathways.* ProgNeurobiol. 2006; 79(4): 173-89. [DOI:10.1016/j.pneurobio.2006.07.006] [PubMed] [Google Scholar]

32. Sadeghipour F, Gharakhanlo R, Rahmati M, Movahhedin M. *The effect of six weeks endurance training on GSK-3β gene expression in the motor spinal cord of male Wistar rats with diabetes neuropathy.* Sport Biosciences. 2018; 9(1): 333-349. [Google Scholar]

33. Dae-Young Kim, Sun-Young Jung, Kijeong Kim, Chang-Ju Kim. *Treadmill exercise ameliorates Alzheimer disease-associated memory loss through the Wnt signaling pathway in the streptozotocin-induced diabetic rats.* Journal of Exercise Rehabilitation 2016; 12(4): 276-283. [DOI:10.12965/jer.1632678.339] [PubMed] [Google Scholar]

34. Stranahan AM, Lee K, Becker KG, Zhang Y, Maudsley S, Martin B, et al. *Hippocampal gene expression patterns underlying the enhancement of memory by running in aged mice.* Neurobiol Aging. 2010; 31: 1937-1949. [DOI:10.1016/j.neurobiolaging.2008.10.016] [PubMed] [Google Scholar]

35. Tiwari SK, Agarwal S, Seth B, Yadav A, Nair S, Bhatnagar P, et al. *Curcumin-loaded nanoparticles potently induce adult neurogenesis and reverse cognitive deficits in Alzheimer's disease model via canonical Wnt/β-catenin pathway*. ACS Nano. 2014; 8: 76-103. [DOI:10.1021/nn405077y] [PubMed] [Google Scholar]

36. Lo GH, LaValley M, McAlindon T, Felson DT. *Intra-articular hyaluronic acid in treatment of knee osteoarthritis:a meta-analysis.* JAMA. 2003; 290: 3115-21. [DOI:10.1001/jama.290.23.3115] [PubMed] [Google Scholar]

37. Ghosh P, Guidolin D. *Potential mechanism of action of intra-articular hyaluronan therapy in osteoarthritis: are theeffects molecular weight dependent?* Semin Arthritis Rheum. 2002; 32: 10-37. [DOI:10.1053/sarh.2002.33720] [PubMed] [Google Scholar]

38. Goldberg VM, Buckwalter JA. *Hyaluronans in the treatment of osteoarthritis of the knee: evidence fordiseasemodifying activity*. Osteoarthritis Cartilage. 2005; 13: 216-24. [DOI:10.1016/j.joca.2004.11.010] [PubMed] [Google Scholar]

39. Listrat V, Ayral X, Patarnello F, Bonvarlet JP, Simonnet J, Amor B, et al. *Arthroscopic evaluation of potentialstructure modifying activity of hyaluronan (Hyalgan) in osteoarthritis of the knee.* Osteoarthritis Cartilage.1997; 5: 153-60. [DOI:10.1016/S1063-4584(97)80010-6] [PubMed] [Google Scholar]

40. Guidolin DD, Ronchetti IP, Lini E, Guerra D, Frizziero L. *Morphological analysis of articular cartilage biopsies from arandomized, clinical study comparing the effects of 500-730 kDasodium hyaluronate (Hyalgan) and methylprednisolone acetate on primary osteoarthritis of the knee.* Osteoarthritis Cartilage. 2001; 9(4): 371-81. [DOI:10.1053/joca.2000.0398] [PubMed] [Google Scholar]

41. Takahashi K, Hashimoto S, Kubo T, Hirasawa Y, Lotz M, Amiel D. *Hyaluronan suppressed nitric oxide production in the meniscus and synovium of rabbit osteoarthritis model.* J Orthop Res. 2001; 19(3): 500-3. [DOI:10.1016/S0736-0266(00)90024-X] [PubMed] [Google Scholar]

42. Díaz-Gallego L, Prieto JG, Coronel P, Gamazo LE, Gimeno M, Alvarez AI. *Apoptosis and nitric oxide in an experimental model of osteoarthritis in rabbit after hyaluronic acid treatment.* J Orthop Res. 2005; 23(6): 1370-6. [DOI:10.1016/j.orthres.2005.05.003.1100230619] [PubMed] [Google Scholar]

How to Cite:

Alinezhad H, Abbassi-Daloii A,Farzanegi P, Abdi A. [Response of Cardiac Tissue β-catenin and GSK-3β to Aerobic Training and Hyaluronic Acid in Knee OA Model Rats]. mljgoums. 2021; 15(1): 45-53. DOI: 10.29252/mlj.15.1.45