HEPATIC AND SKELETAL MUSCLE TISSUE IMMUNOMETABOLISM IN TRAINED RATS

IMUNOMETABOLISMO TECIDUAL HEPÁTICO E MUSCULAR ESQUELÉTICO DE RATOS TREINADOS

INMUNOMETABOLISMO DEL TEJIDO HEPÁTICO Y MUSCULAR ESQUELÉTICO DE RATAS ENTRENADAS

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ABSTRACT

Introduction: Physical exercise can be an alternative for preventing and treating the harmful effects of obesity, mainly inflammatory effects on skeletal muscle and liver tissues. However, no consensus exists regarding this purpose's best physical training model. Objective: Evaluate morphological, metabolic, and inflammatory alterations in rats' skeletal and hepatic muscle tissues caused by aerobic and resistance training. Methods: 24 Wistar rats were divided into sedentary (S), aerobic (AE), and resistance training (R) groups. Blood glucose, total cholesterol, and serum triglycerides were measured periodically. After euthanasia, body mass was measured to calculate the total mass gain during the experiment. High-density lipoprotein (HDL) was measured. Adipose tissue was extracted to calculate its percentage relative to body mass and the liver, soleus, and gastrocnemius muscles for morphological analyses and concentrations of glycogen, lipids, and Tumor Necrosis Factor α (TNF-α). The Kruskall-Wallis test and Dunn's post-test were performed for statistical analysis, adopting $p<0.05$. Results: Both training models reduced the percentage of adipose tissue, body mass gain, and hepatic TNF-α concentration ($p<0.05$). AE increased serum HDL, gastrocnemius fiber diameter and reduced the fractal dimension in the soleus ($p<0.05$). R reduced blood glucose and serum and liver lipids, increased liver and soleus glycogen concentrations, increased gastrocnemius fiber diameter, and decreased TNF-α ($p<0.05$). Conclusion: Both training models reduced body mass, relative visceral adipose tissue, serum total cholesterol concentration, and liver inflammation. However, resistance training was more effective in promoting metabolic effects in the liver and skeletal muscle and reducing muscle inflammation in rats. ***Level of Evidence V; Expert Opinion.***

Keywords: Resistance Training; Liver; Muscle, Skeletal; Tumor Necrosis Factor-alpha.

RESUMO

Introdução: O exercício físico pode se apresentar como uma alternativa para prevenção e tratamento de efeitos deletérios da obesidade, principalmente efeitos inflamatórios sobre os tecidos muscular esquelético e hepático. No entanto, não há consenso quanto ao melhor modelo de treinamento físico para tal finalidade. Objetivos: Avaliar alterações morfológicas, metabólicas e inflamatórias dos treinamentos aeróbico e resistido sobre os tecidos muscular esquelético e hepático de ratos. Métodos: 24 ratos Wistar foram divididos nos grupos sedentário (S), treinamento aeróbico (AE) e resistido (R). Glicemia, colesterol total e triglicerídeos séricos foram mensurados periodicamente. Após a eutanásia, a massa corporal foi mensurada para calcular o ganho total de massa durante o experimento. A lipoproteína de alta densidade (HDL) foi dosada. O tecido adiposo foi extraído para cálculo de sua porcentagem relativa à massa corporal assim como o fígado e os músculos sóleo e gastrocnêmio para as análises morfológicas e das concentrações de glicogênio, lipídios e Fator de Necrose Tumoral a (TNF-α). Para análise estatística, foram utilizados o teste de Kruskall-Wallis e o pós-teste de Dunn, adotando-se $p<0,05$. Resultados: Ambos os modelos de treinamento reduziram o percentual de tecido adiposo, ganho de massa corporal e concentração hepática de TNF-α ($p<0,05$). AE aumentou o HDL sérico, o diâmetro das fibras do gastrocnêmio e reduziu a dimensão fractal no sóleo ($p<0,05$). R reduziu a glicemia e os lipídios séricos e hepáticos, aumentou a concentração de glicogênio hepático e sóleo, aumentou o diâmetro das fibras gastrocnêmicas e diminuiu o TNF-α ($p<0,05$). Conclusão: Ambos os modelos de treinamento reduziram a massa corporal, o tecido adiposo visceral relativo, a concentração sérica de colesterol total e a inflamação hepática. No entanto, o treinamento resistido mostrou-se mais eficaz em promover efeitos metabólicos no fígado e no músculo esquelético, além de reduzir a inflamação muscular em ratos. ***Nível de Evidência V; Opinião do Especialista.***

Descritores: *Treinamento de Resistência; Fígado; Músculo Esquelético; Fator de Necrose Tumoral alfa.*

RESUMEN

Introducción: El ejercicio físico puede ser una alternativa para prevenir y tratar los efectos nocivos de la obesidad, principalmente los efectos inflamatorios sobre los tejidos del músculo esquelético y del hígado. Sin embargo, no existe consenso sobre cuál es el mejor modelo de entrenamiento físico para este fin. Objetivo: Evaluar las alteraciones morfológicas, metabólicas e inflamatorias del entrenamiento aeróbico y de resistencia en sobre los tejidos músculo esqueléticos y hepáticos de ratas. Métodos: 24 ratas Wistar se dividieron en grupos sedentarios (S), aeróbicos (AE) y

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de entrenamiento de resistencia (R). Se midieron periódicamente glucosa en sangre, colesterol total y triglicéridos. Después de la eutanasia, se midió la masa corporal para calcular la ganancia de masa total durante el experimento. Se midió la lipoproteína de alta densidad (HDL). Se extrajo tejido adiposo para calcular su porcentaje relativo a la masa corporal, así como hígado, músculos sóleo y gastrocnemio para análisis morfológicos y concentraciones de glucógeno, lípidos y Factor de Necrosis Tumoral α (TNF-α). Para el análisis estadístico fueron utilizados Kruskall-Wallis y el post-test de Dunn, adoptando p<0,05. Resultados: Ambos entrenamientos redujeron el porcentaje de tejido adiposo, masa corporal y la concentración de TNF-α hepático (p<0,05). AE aumentó el HDL sérico, el diámetro de la fibra del gastrocnemio y redujo la dimensión fractal en el sóleo (p<0,05). R redujo la glucosa en sangre y los lípidos séricos y hepáticos, aumentó las concentraciones de glucógeno hepático y sóleo, aumentó el diámetro de la fibra del gastrocnemio y disminuyó el TNF-α (p<0,05). Conclusión: Ambos modelos de entrenamiento redujeron la masa corporal, el tejido adiposo visceral relativo, la concentración sérica de colesterol total y la inflamación hepática. El entrenamiento de resistencia demostró ser más eficaz para promover los efectos metabólicos en el hígado y el músculo esquelético, además de reducir la inflamación muscular en ratas. ***Nivel de Evidencia V; Opinión del Especialista.***

Descriptores: *Entrenamiento de Resistencia, Hígado; Músculo Esquelético; Factor de Necrosis Tumoral alfa.*

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INTRODUCTION

Due to its subtle symptoms and long-term effects, sedentary behavior remains a popular topic. A sedentary lifestyle can create a positive feedback loop: reduced physical capacity leads to discomfort during exercise, which can discourage further activity.[1]

As a result, body fat accumulates, caused by an imbalance between calorie intake and energy expenditure. This increase can lead to other pathological conditions, such as metabolic syndrome. In addition, hypertrophied adipocytes suffer hypoxia and secrete Tumor Necrosis Factor α (TNF-α), responsible for the onset of chronic diseases.[2]

However, physical training can be an effective non-pharmacological alternative to these harmful effects. Both aerobic and resistance training can reduce the risk of developing chronic diseases,[3,4] since muscle contraction stimulates the production of Interleukin 10 and reduces the secretion of TNF-α.[5,6]

However, it is unclear which type of training is best suited to reducing the effects of a sedentary lifestyle and whether the anti-inflammatory effect promoted occurs similarly in skeletal muscle and liver tissues. Therefore, the present study evaluated the morphological, metabolic, and inflammatory changes that aerobic and resistance training promoted in rats' skeletal muscle and liver tissue.

MATERIALS AND METHODS

Animals

Twenty-four seven-week-old male Wistar rats were obtained from the Animal House II of the Faculty of Science and Technology of the Universidade Estadual Paulista "Júlio de Mesquita Filho" (FCT/UNESP), Presidente Prudente campus. The research was carried out by the rules and ethical principles of animal experimentation of the Ethics Committee for the Use of Animals (CEUA) of FCT/UNESP, under protocol No. 003/2016.

Experimental groups

The animals were divided into sedentary (S), aerobic (AE), and resistance (R) groups and fed SupraLab® brand rodent food from Alisul® (São Leopoldo, Brazil), with a composition of 48g of carbohydrates, 25g of proteins, 3g of lipids and 24g of fibers and minerals per 100 grams, totaling 2.95 kilocalories per gram (Kcal/g). Tap water was offered *ad libitum* for hydration.

Training protocols

Protocol for the sedentary group

Group S remained in the cage for the entire experimental period without any experimental physical training.

Aerobic training protocol

To familiarize themselves with aerobic training, the animals walked on the Inbramed® treadmill for 30 minutes once a day for six consecutive days, according to the method proposed by Lira et al.[7] After the familiarization period, the prescription of the loads used in aerobic training was defined by the Maximum Speed Test (V_{max}), using the methods of Faleiros et al.[8]

Then the training period began, in which the animals ran on a treadmill for 60 minutes, once a day, five times a week, for 12 weeks, following the model proposed by Lira et al.[7]

Resistance training protocol

Resistance training was carried out in an aquatic environment. A cylindrical plastic container with 50 cm high and 25 cm in diameter was adapted. The container was filled with water at an average temperature of 30 ± 2 ºC until it reached a depth of 38 cm. The workload was accommodated in the posterior region of the chest using a special vest, allowing vertical jumps to the surface of the container. The familiarization period was carried out as proposed by Teixeira et al.[9]

After the familiarization period, the resistance training protocol began, consisting of four sets of 10 jumps, with a 60-second interval between sets, three times a week, for 12 weeks, following the model used by Teixeira et al.[9]

Periodic evaluation

The animals were periodically assessed for blood glucose and lipid profile during the experimental period, starting one week before the beginning of the pre-training periods, the 8th and 13th weeks of the experimental period.

Analysis of the glycemic response during the experimental period

Blood samples of 25µl were collected from the distal end of the animals' tails for analysis by photometry using the Accu-Check Active® digital monitor (Roche Diagnosis®, Basel, Switzerland). In the pre-training period, only resting blood glucose was collected from the animals in all groups. On the 13th week, a blood glucose test was carried out on the animals in all groups, as proposed by Oliveira et al.[10]

Analysis of the lipid profile during the experimental period

The lipid profile was assessed pre-training on the 8th and 13th weeks of training. New blood samples of 25µl were collected from the distal end of the animals' tails for photometric analysis of total cholesterol and triglycerides using the Accutrend Plus® monitor from Roche Diagnostics® (Basel, Switzerland). This monitor measured triglycerides from 69 mg/dl and total cholesterol from 149 mg/dl. Thus, total cholesterol values

were categorized as higher or lower than 149 mg/dl. Triglyceride values were presented in absolute numbers.[11]

Euthanasia

Body mass was determined using an Ohaus® analytical scale to calculate the animals' mass gain during the experimental period (Initial Mass - Final Mass = Body Mass Gain). The animals were anesthetized and euthanized according to the methods of Lira et al.[7] 10ml of blood samples were collected to measure High-Density Lipoprotein (HDL) cholesterol. A paramedial incision was made in the abdomen, with the epithelium being folded back to collect the epididymal, mesenteric, and retroperitoneal adipose tissues and the liver. Another incision was made on the lateral side of the tibial region of the lower limb to collect the soleus and gastrocnemius muscles.[12]

Additional variables for the lipid profile

Measurement of relative adipose tissue

The epididymal, mesenteric, and retroperitoneal adipose tissues were weighed on a precision scale, and the percentage relative to the animal's body mass (%) was calculated.[13]

HDL cholesterol measurement

The blood samples were used to measure HDL cholesterol using enzymatic methods (CENTERKIT®, São Paulo, Brazil), and colorimetric density was read using a 1600UV® spectrophotometer, brand BUNKER® (Piracicaba, Brazil), with a wavelength of 600nm.[7]

Histological preparation and stereological analysis of liver tissue

Histological preparation of the liver tissue was carried out using the methods of Barbosa Magalhães et al.[13] All the images for histological analysis were obtained using a Nikon® 50i optical microscope (New York, United States), coupled with a Nikon® Infinity 1 digital camera (New York, United States).

Ten images per animal were used at 400x magnification for the stereological analysis and stained with Hematoxylin and Eosin (HE). ImageJ® 1.47t for Windows® software (National Institute of Health - Maryland, United States) was used to apply the 144-point multipurpose test recommended by Weibel.[14] Using the methods of Catta-Preta et al.,[15] the relative cellular, nuclear, and extracellular matrix portions and the cellular numerical density were obtained.

Histological preparation and morphometric and fractal dimension analysis of skeletal muscle tissue

The soleus and gastrocnemius muscles underwent the histological preparation recommended by Camargo et al.[12] The histological slides of the soleus muscle were stained with HE,[12] PAS[16] and Sudam III.[16] Among the histological slides of the gastrocnemius muscle, one sample was stained with HE, and another was immunolocalized with TNF-α antibodies.

Morphological analysis was conducted using NIS-Elements D 3.0 - SP7 - build 547 software, Nikon® for Windows® (New York, United States). In the images of samples stained with HE and magnified at 100x, the smallest diameter of 100 fibers of the soleus and gastrocnemius muscles of each animal was measured.[12]

The Fractal Dimension of the soleus and gastrocnemius muscles was analyzed using ImageJ® 1.47t for Windows® software (National Institute of Health - Maryland, United States), using the systems proposed by Moreira et al.[17] and Ozaki et al.[18]

Quantification of concentration of glycogen, lipids, and TNF-α in liver and skeletal muscle tissues

Periodic Acid Schiff (PAS)[16] was used to assess tissue glycogen concentration, and Sudam III stain[19] was used to assess lipid concentration. In addition, the Tumor Necrosis Factor Alpha (TNF-α) antibody was immunolocalized.

These analyses used images at 20x magnification for skeletal muscle tissue samples and 40x magnification for liver tissue, using ImageJ® 1.47t for Windows® software (National Institute of Health - Maryland, United States), using the method proposed by Da Rocha et al.[19]

Statistical Analysis

Sample sizing was used to calculate the minimum number of animals needed, resulting in eight animals per group; the Randomized Block Design in Factorial Scheme was used to randomize the animals.[20] The Wilcoxon test defined the prevalence of total cholesterol.[20] The Shapiro--Wilk test was used to verify the normality of the data, and the statistical analysis was carried out using the Kruskall-Wallis test and the post-test of Dunn,[20] adopting $p<0.05$ and using the IBM SPSS 22.0® software.

RESULTS

Table 1 shows no significant difference between the initial (g) and final (g) body mass groups. Still, there was a reduction in body mass in groups AE (p=0.009) and R (p=0.001) compared to S during the experimental period. Serum triglyceride levels in the R group were lower than in the S group at week 13 (p=0.025), with no significant difference in the other periods.

The adipose tissue mass (g) in group S was higher than in group R (p=0.039). On the other hand, the relative adipose tissue (%) of the S group was higher compared to the AE (p=0.037) and R (p=0.039) groups. Finally, group R had a lower glycemic concentration than group S (p=0.044) at the end of the test (25th minute).

Table 2 shows a higher prevalence of total cholesterol above 149 mg/dl in group S (p=0.034). Serum HDL cholesterol levels (mg/dl) in the AE group were higher than in the S group (p=0.021).

Liver tissue analysis

Table 3 shows that groups AE and R had a lower numerical density of hepatocytes (p=0.001 and p=0.028), a lower cell volume (p=0.001), and

Table 1. Values referring to body mass (g), serum triglyceride concentration (TG, in mg/dl), mass (g) and percentage (%) of adipose tissue and glycemic test after the experiment.

Variables (Med (IQ))	PS (n=8)	AE (n=7)	R (n=6)
Initial body mass (g)	336.5 (29.37)	343.5 (28.00)	352.5 (66.12)
Final body mass (g)	474.2 (62.00)	463.5 (46.50)	434.0 (91.25)
Body mass gain (g)	156.5 (34.25)	124.0 (43.00) *	100.5 (65.50) *
Pre-training TG (mg/dl)	101.5 (27.25)	108.0 (41.00)	136.0 (31.50)
TG 8th week (mg/dl)	127.5 (23.50)	118.0 (60.50)	114.0 (32.50)
TG 13th week (mg/dl)	132.5 (26.50)	125.0 (69.50)	114.0 (32.50) *
Mass of adipose tissue (g)	15.52 (5.08)	12.29 (4.55)	9.38 (6.51) *
Relative adipose tissue (%)	3.54 (0.81)	3.05 (0.83) *	2.48 (1.21) *
Resting blood glucose (mg/dl)	89.00 (10.00)	116.00 (10.00)	132.00 (30.00)
Blood glucose 5th min test (mg/dl)	91.00 (4.00)	103.00 (24.00)	136.00 (48.00)
Blood glucose 25th min test (mg/dl)	90.00 (16.00)	102.00 (26.50)	108.00 (31.75) *

Caption: S = Sedentary; AE = Aerobic; R = Resistance; Med = Median; IQ = Interquartile Range; TG = triglycerides. Kruskall-Wallis test and Dunn's post-test to compare results. * $p < 0.05$ compared to PS.

Table 2. Prevalence (%) of animals with total cholesterol above 149 mg/dl during the experimental period and HDL cholesterol values (mg/dl) after euthanasia.

	Total cholesterol (%)			HDL (Med (IQ))
	Pre-training	Week 8	Week 13	Post-euthanasia
PS	50.0%	37.5%	87.5%*	35.36 (8.10)
PAE	42.9%	85.7%	14.3%	42.71 (8.09) *
PR	67.7%	33.3%	50.0%	42.97 (18.72)

Caption: S = Sedentary; AE = Aerobic; R = Resistance; Med = Median; IQ = Interquartile Range. Wilcoxon test to compare total cholesterol results. Kruskall-Wallis test and Dunn's post-test to compare HDL cholesterol results. * p<0.05 compared to PS.

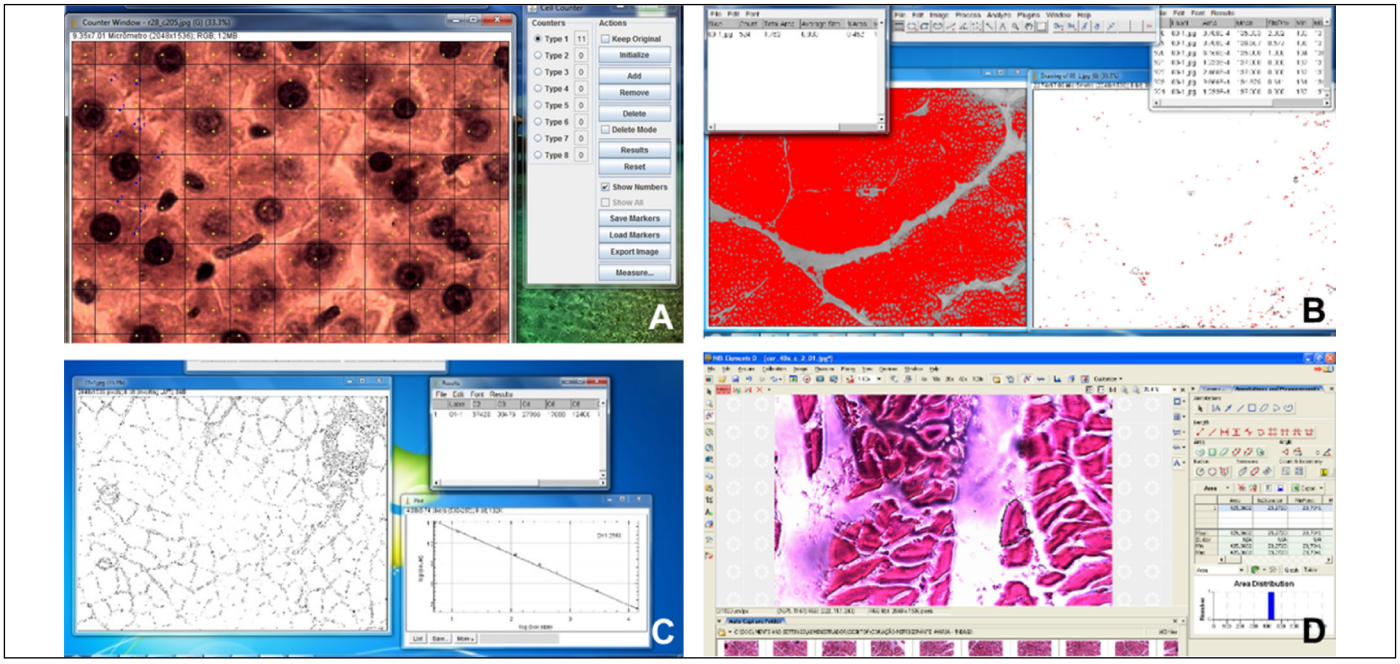

Figure 1. Use of software: A, B, and C. ImageJ® 1.47t for Windows® for stereological analysis, estimation of glycogen, lipid, IL-6, and TNF-α concentrations and fractal dimension analysis in liver and skeletal muscle tissues; D. NIS-Elements D 3.0 - SP7 - build 547, Nikon® for Windows® for measuring the smallest diameter of muscle fibers. (Source: LAPMUS).

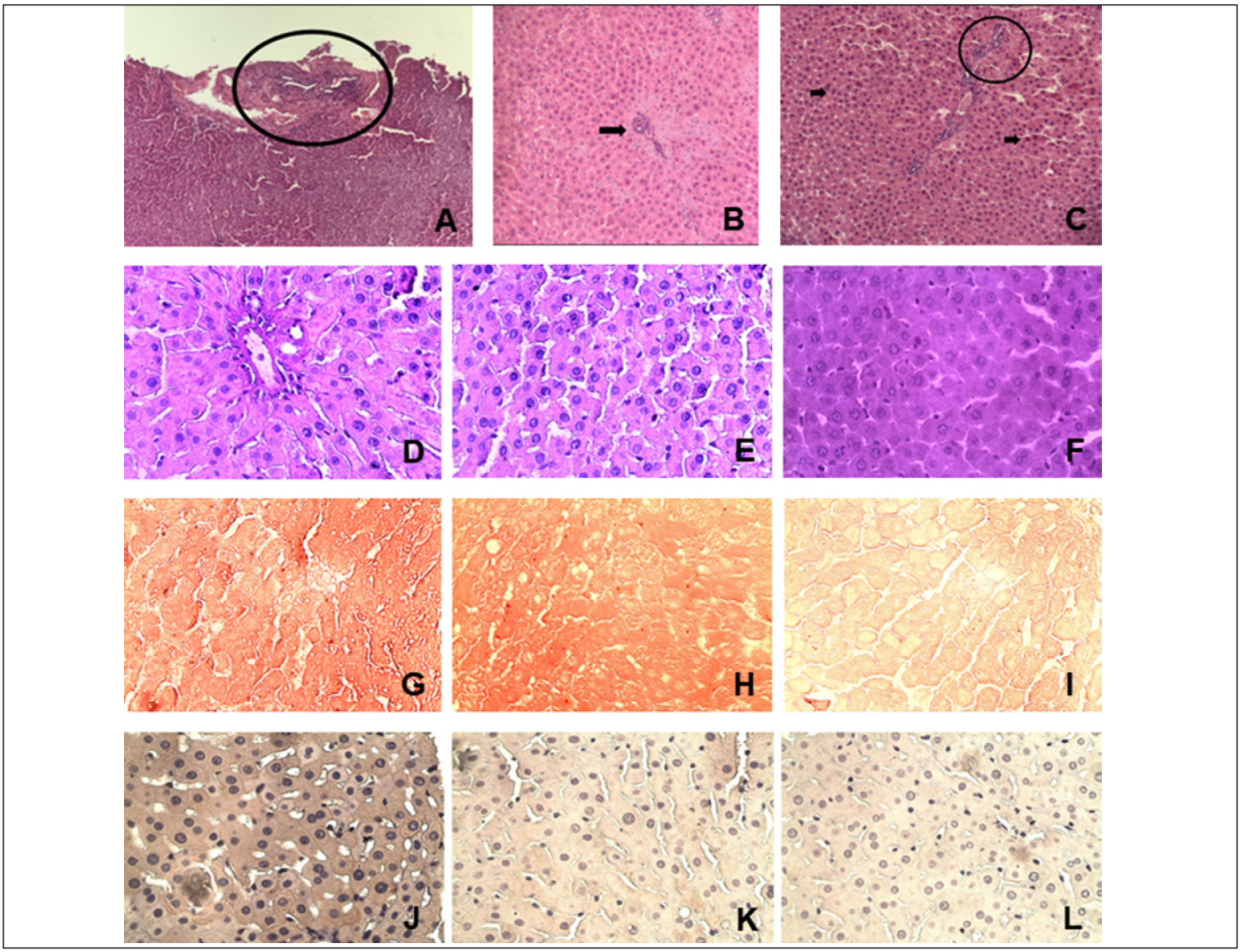

Figure 2. Photomicrographs of liver tissue: A. PS with a thickness of 5µm and HE staining. Moderate lymphocyte increase in portal spaces (circle) without lymphocyte extravasation, 200X; B. PAE, 5µm thick and HE stained. Discrete lymphocyte increase, without structural alterations or activity (arrow), 200X; C. PR, 5µm thick and HE stained. Moderate lymphocyte increase in portal spaces (circle) with extravasation of lymphocytes and frequent binucleated hepatocytes, some with pyknotic nuclei (arrows). 200X; D. PS with 5µm thickness and PAS staining, 400X; E. PAE; F. PR; G. PS with 5µm thickness and Sudam III staining, 400X; H. PAE; I. PR; J. PS with 5µm thickness and immunolocalized by TNF-α antibody, 400X; K. PAE; L. PR.

Table 3. Stereological analysis values and concentrations (%) of glycogen, lipids, and TNF-α in liver tissue after the experimental period.

Variables (Med (IQ))	S (n=8)	AE (n=7)	R (n=6)
Liver mass (g)	11.69 (1.38)	12.70 (2.71)	10.46 (1.42)
Numerical density (nº/μm³)	1.03 (0.05)	0.84 (0.26) *	0.87 (0.23) *
Cell volume (%)	84.72 (2.43)	79.16 (7.63) *	56.25 (10.06) *
Nuclear volume (%)	14.58 (5.20)	26.38 (12.84)	21.52 (9.37)
Volume of extracellular matrix (%)	15.27 (2.43)	20.83 (7.63) *	43.75 (10.06) *
Glycogen (%)	27.70 (16.46)	38.49 (22.55)	47.53 (16.49) *
Lipids (%)	54.62 (14.41)	51.74 (17.57)	38.55 (25.58) *
TNF-α (%)	40.53 (6.76)	22.44 (7.79) *	21.29 (16.11) *

Caption: S = Sedentary; AE = Aerobic; R = Resistance; Med = Median; IQ = Interquartile Range. Kruskall-Wallis test and Dunn's post-test to compare results. * p<0.05 compared to S.

a higher extracellular matrix (p=0.001) compared to S in the sterological analysis, with no significant difference in terms of liver mass.

The R group had a higher concentration of glycogen (p=0.001) and a lower concentration of lipids (p=0.003) compared to the S. In addition, there was a lower concentration of TNF-α in the AE and R groups (p=0.001) compared to the S, with no significant difference in the concentration of IL-6.

Analysis of skeletal muscle tissue

Table 4 shows no difference in the mass and diameter of the soleus muscle fibers, but AE had a lower fractal dimension than S and R (p=0.001). Group R had a higher muscle glycogen concentration than AE (p=0.001), with no significant difference in lipid concentration.

In the gastrocnemius muscle, there was no difference in muscle mass between the groups, but AE (p=0.001) and R (p=0.003) had larger

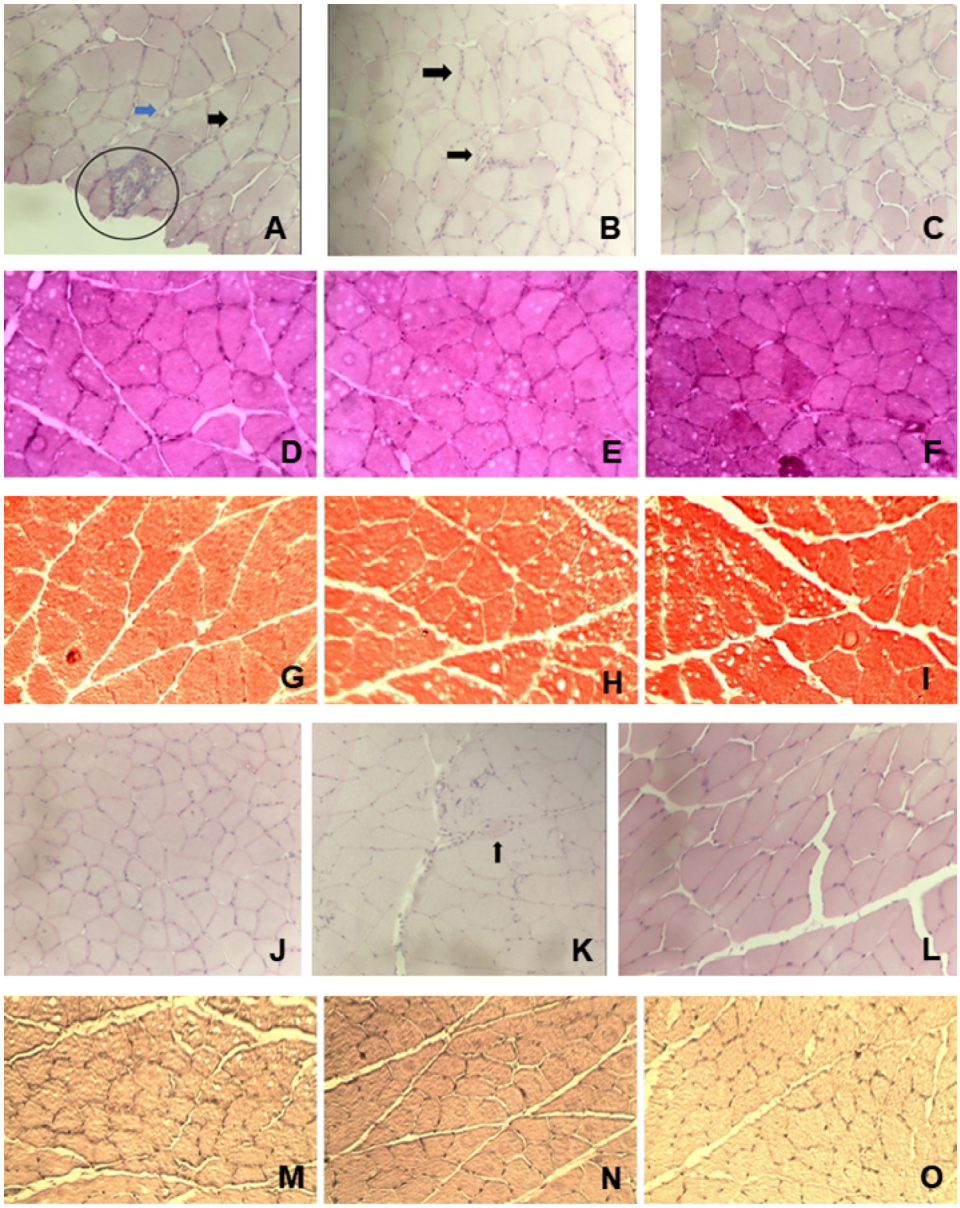

Figure 3. Photomicrographs of soleus skeletal muscle tissue: A. PS, 5μm thick and HE stained. Thickening of the perimysium (blue arrow) and endomysium (black arrow) with lymphohistiocytic infiltrate attacking muscle cells (circle), 200X; B. PAE, 5μm thick and HE stained. PAE, 5μm thick and HE stained. Endomysium focally thickened and presence of discreet inflammatory infiltrate (arrows), 200X; C. PR, 5μm thick and HE stained. No significant changes in the tissue, 200X. D. PS, 5μm thick and stained with PAS, 200X; E. PAE; F. PR; G. PS, 5μm thick and stained with Sudam III, 200X; H. PAE; I. PR; J. Photomicrograph of the skeletal muscle tissue of the gastrocnemius of the PS subgroup, with a thickness of 5μm and HE staining. There are no significant histological changes. Isomorphic polygonal skeletal muscle cells with preserved perimysial and endomysial contours, 200X; K. PAE, 5μm thick and HE stained. Focal perimysial thickening with rare lymphocytes (arrow), 200X; L. PR, 5μm thick and HE stained. There are no significant histological changes. Isomorphic polygonal skeletal muscle cells with preserved perimysial and endomysial contours, 200X; M. PS, 5μm thick and immunolocalized by TNF-α antibody, 200X; N. PAE; O. PR.

Table 4. Values of the smallest diameter of muscle fibers (μm), fractal dimension analysis (UA) and concentrations (%) of glycogen, lipids and TNF-α in the soleus and gastrocnemius muscles after the experimental period.

Variables (Med (IQ))	S (n=8)	AE (n=7)	R (n=6)
SL mass (g)	0.103 (0.02)	0.099 (0.01)	0.097 (0.01)
GT mass (g)	0.610 (0.07)	0.568 (0.24)	0.655 (0.10)
Smallest SL diameter (μm)	71.34 (9.89)	64.52 (6.56)	64.44 (9.88)
Smallest GT diameter (μm)	55.06 (7.03)	70.05 (5.70) *	70.38 (11.66) *
DF SL (UA)	1.586 (0.06)	1.510 (0.11) * †	1.558 (0.07)
DF GT (UA)	1.664 (0.04)	1.532 (0.08)	1.565 (0.12) * **
Glycogen SL (%)	47.55 (13.88)	34.08 (18.40)	57.57 (36.11) **
Lipids SL (%)	68.30 (17.48)	64.23 (11.87)	75.19 (18.51)
TNF-α GT (%)	22.72 (13.95)	24.25 (9.76)	14.50 (10.33) * **

Caption: S = Sedentary; AE = Aerobic; R = Resisted; Med = Median; IQ = Interquartile Range; FD = Fractal Dimension; SL = soleus; GT = gastrocnemius; UA = arbitrary unit. Kruskall-Wallis test and Dunn's post-test to compare results. * p<0.05 compared to S. ** AE. † R.

muscle fiber diameters compared to S. Group R had lower fractal dimension values compared to S and AE (p=0.001). There was no difference in the concentration of IL-6, but R had a lower concentration of TNF-α compared to S and AE (p=0.001).

DISCUSSION

The AE group had lower total cholesterol and HDL concentrations than the S group. According to Rahmati-Ahmadabad et al.,[21] aerobic training increases HDL formation by increasing the hepatic expression of Intracellular Lipid Transporters ATP-binding cassette 1 (ABC1), increasing the transport of total cholesterol and reducing its serum concentration.

In addition, the reduction in visceral adipose tissue and body mass in the AE group compared to the S group may be supported by the transient pro-oxidative state in white adipose tissue after training, favoring increased lipolysis and reduced lipogenesis in adipocytes.[22]

The increase in hepatic and muscular glycogen deposits and lower glycemic concentration presented by R compared to S may have occurred due to the increase in insulin sensitivity, which results in absorption and cellular glycolysis after training.[23]

At the same time, there was a reduction in body mass, visceral adipose tissue, hepatic lipid concentrations, and serum concentrations of triglycerides and total cholesterol in the R group. Domingos et al.[24] observed that resistance training reduces the hepatic expression of SREBP1c and Stearyl-CoA-Desaturase 1 (SDC-1). Thus, post-workout neoglycogenesis occurs from free fatty acids,[23] which reduces hepatic lipogenesis, increases lipolysis in adipocytes, and reduces serum triglyceride concentrations.[24]

The higher concentration of TNF-α and the stereological values of the S group compared to the AE and PR groups suggest an inflammatory process and tissue alterations resulting from a sedentary lifestyle.[23-27] Previous studies[3,4] have observed a reduction in hepatic IL-1β expression due to aerobic training. Resistance training directly inhibits the transcription of NF-κB and reduces the synthesis of hepatic TNF-α, which may explain the changes in the liver tissue of sedentary animals compared to trained ones.[3,4]

The AE group showed lower fractal dimension values in the soleus muscle than the others, possibly related to lower muscle tissue inflammation.[18] Kim et al.,[28] observed an increase in the production of Uncoupling Protein 2 (UCP-2) followed by a reduction in the concentration of TNF-α in the soleus muscle after aerobic training. These findings suggest that aerobic training reduces inflammation in the soleus muscle of EA.

Both trained groups had larger gastrocnemius muscle fiber diameters. Resistance training is known to inhibit the Transforming Growth Factor β1 (TGF-β1) signaling pathway,[29] while aerobic training activates the Insulin-Linked Growth Factor 1 (IGF-1) and Mammalian Target of Rapamycin (mTOR) signaling pathways.[30] Both pathways promote satellite cell activation, post-training protein synthesis for tissue regeneration, and increased muscle trophism in the gastrocnemius.

The lower value in the fractal dimension analysis and the lower concentration of TNF-α in the gastrocnemius of group R compared to the others indicate a reduction in tissue inflammation since resistance training increases the expression of PGC-1α, which activates muscle oxidative capacity and inhibits TNF-α synthesis.[5]

However, there was a reduction in muscle inflammation in the soleus of the AE group and the gastrocnemius of the R group. Lira et al.,[7] found that the anti-inflammatory effect in muscles with a predominance of glycolytic metabolism may be related to glycogen content and the physical stress caused by resistance training. In contrast, in muscles with a predominance of aerobic metabolism, the anti-inflammatory effect is related to muscle oxidative capacity during aerobic training. These findings may justify the anti-inflammatory effects on the soleus and gastrocnemius muscles.

CONCLUSION

Both training models reduced body mass, relative visceral adipose tissue, serum total cholesterol concentration, and liver inflammation. However, resistance training promoted better metabolic effects in the liver and skeletal muscle and reduced muscle inflammation in rats compared to aerobic training.

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