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Original Article

EFFECT OF A SINGLE RESISTANCE EXERCISE SESSION ON INFLAMMATORY MARKERS AND OXIDATIVE STRESS IN WOMEN LIVING WITH HIV

EFEITO DE UMA ÚNICA SESSÃO DE EXERCÍCIO RESISTIDO SOBRE MARCADORES INFLAMATÓRIOS E ESTRESSE OXIDATIVO EM MULHERES VIVENDO COM HIV

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RESUMO

Pessoas vivendo com HIV (PVH), que utilizam a terapia antirretroviral (TARV), são mais suscetíveis a alterações no perfil inflamatório e estresse oxidativo, sendo que as mulheres possuem maior acesso à TARV. Embora o exercício físico seja uma estratégia complementar ao tratamento devido aos seus efeitos antioxidantes e anti-inflamatórios, não está claro se as respostas agudas ao exercício podem ser prejudiciais às PVH. O objetivo do estudo foi investigar o efeito agudo de exercícios com pesos (EP) sobre marcadores inflamatórios e de estresse oxidativo em PVH. Dez mulheres, em uso da TARV, realizaram uma sessão de EP constituída por sete exercícios para o corpo todo. Para avaliação bioquímica, amostras de sangue foram coletadas antes (pré), 1 hora (1h) e 2 horas (2h) após a sessão de EP. A ANOVA *one-way* seguida do teste *post hoc* de Bonferroni foi utilizada para comparação dos resultados entre os momentos. Houve aumento apenas nos marcadores, GSSG de 160% (pré: 0,40 ± 0,11; 1h: 1,18 ± 0,36; 2h:1,04 ± 0,25 mmol/g), TNF-α de 98% (pré: 4,60 ± 0,55; 1h: 6,95 ± 0,77; 2h: 9,10 ± 1,03 pg/ml) e IL-6 de 52% (pré: 2,47 ± 0,67; 1h: 3,63 ± 1,26; 2h: 5,38 ± 2,15 pg/ml). As demais variáveis permaneceram sem alterações (P > 0,05). Conclui-se que uma sessão de EP aumentou os níveis de marcadores inflamatórios e estresse oxidativo em PVH de forma não exacerbada.
Palavras-chave: Infecções por HIV. Citocinas. Radicais livres. Exercício agudo.

ABSTRACT

People living with HIV (PLH), who use antiretroviral therapy (ART), are more susceptible to changes in the inflammatory profile and oxidative stress, and women have greater access to ART. Although physical exercise is a complementary strategy to treatment due to its antioxidant and anti-inflammatory effects, it is not clear whether acute responses to exercise can be harmful to PLH. The aim of the study was to investigate the acute effect of resistance exercise (RE) on inflammatory and oxidative stress markers in PLH. Ten women, using ART, performed RE session consisting of seven exercises for the whole body. For biochemical evaluation, blood samples were collected before (pre), 1 hour (1h) and 2 hours (2h) after the RE session. One-way ANOVA followed by Bonferroni's post hoc test was used to compare results between time points. There was an increase only in markers, GSSG of 160% (pre: 0.40 ± 0.11; 1h: 1.18 ± 0.36; 2h: 1.04 ± 0.25 mmol/g), TNF-α of 98 % (pre: 4.60 ± 0.55; 1h: 6.95 ± 0.77; 2h: 9.10 ± 1.03 pg/ml) and 52% IL-6 (pre: 2.47 ± 0 .67; 1h: 3.63 ± 1.26; 2h: 5.38 ± 2.15 pg/ml). The other variables remained unchanged (P > 0.05). It is concluded that a RE session increased the levels of inflammatory markers and oxidative stress in PLH in a non-exacerbated way.
Keywords: HIV Infection. Cytokines. Free Radicals. Acute Exercise.

Introduction

The global prevalence of Human Immunodeficiency Virus (HIV) is estimated at 38.4 million cases[1]. Antiretroviral therapy (ART) has emerged as an effective approach to mitigate morbidity and increase the longevity of HIV-infected individuals[2] effectively transforming HIV into a chronic, manageable disease state[3]. As such, ART has become a mainstay of HIV treatment and care. However, data indicates that adult women have greater access (80%) to ART compared to adult men (70%)[1]. There are reports in the literature that ART can increase

oxidative stress[4,5] and further aggravate the inflammatory status of patients[6]. People living with HIV (PLH) show signs of inflammation with a marked increase in the production of inflammatory cytokines when compared to uninfected individuals[7], in addition to increased production of reactive oxygen species and reduced antioxidant capacity, causing a disorder in antioxidant system[8].

Considering these changes, resistance training has been demonstrated with a non-drug strategy in HIV patients, providing improvement in several parameters[9-13], including anti-inflammatory[14] and antioxidant[15]. Acute exercise creates an inflammatory environment responsible for recovery and adaptation from an exercise session in healthy individuals[16], but it is not known whether the magnitude of this damage is more exacerbated in HIV-infected individuals. Furthermore, there are few studies in the literature that aim to investigate the effects of resistance training on oxidative stress[17,18] and inflammatory markers in PLH[19]. Therefore, this study aims to investigate the effect of a single resistance exercise (RE) session on inflammatory markers and oxidative stress in women living with HIV.

Methods

Participants

The participants included ten women living with HIV, from the Testing and Counseling Center, responsible for the specialized STD/AIDS service in the city of Maringa (Brazil). As inclusion criteria, they must be over 18 years of age; have regularly used ART for more than six months; have a stabilized clinical grade and viral load; have not participated in physical training programs during the previous six months; have no acute or chronic inflammations that could affect physical exercise; and not be pregnant. Individuals who met the inclusion criteria received clinical evaluation by the attending physician based on each patient's history, laboratory tests, and clinical condition.

The study was approved by the Standing Committee on Ethical Research with Humans Beings of the State University of Maringa, PR, Brazil (Process No. 1.245.413). The volunteers signed the Free Informed Consent Form, after being informed about the study proposal and the procedures to which they would be submitted.

Experimental Design

The study adopted a quasi-experimental design. Participants who met the criteria were invited to participate in the familiarization protocol consisting of three sessions with a 48-hour interval between them, with the goal of learning the exercises without the insertion of loads.

The OMNI Resistance Exercise Scale (OMNI-RES) of subjective perceived exertion was used to determine and control the loads for each exercise[20.] The loads used during the experimental sessions corresponded to the intensity equivalent to the five to seven (5-7) range of the OMNI-RES Scale. To achieve this goal, before the beginning of the familiarization with the exercises, anchoring of the OMNI-RES scale was performed. This procedure consists of placing the patient at the lowest and highest load possible in each exercise so that she can differentiate between a lower and higher subjective perception of effort.

After the familiarization sessions, a single exercise session was performed with assessments before and one hour and two hours later.

Biochemical Analyses

Blood samples were collected before (pre), 1 (post 1hr) and 2 hours (post 2hr) after the end of the exercise session. 10 ml of fasting venous blood was collected from everyone in vacutainer® tubes (BD, São Paulo, Brazil), with EDTA. Part of the blood was centrifuged at 3500 rpm for 10 min at 4 ºC. Plasma and whole blood were stored at -80ºC for later analysis.

The viral load was tested using Abbott RealTime HIV-1 Assay. The CD4+ T and CD8+ cell counts were performed by flow cytometry (FACS Calibur Becton-Dickinson, Franklin Lakes, NJ, USA).

The determination of oxidative stress markers occurred through the following analyses: Total hydroperoxide (FOX) was determined according to that described by Södergren et al. (1998)[21]. The determination of advanced oxidation protein products (AOPP) occurred by the protocol suggested by Witko-Sarsat et al. (1996)[22]. The lipoperoxidation products were detected by TBARS determination, as described by Costa et al. (2006)23. Reduced glutathione (GSH), glutathione oxidized (GSSG) and the GSH/GSSG ratio were determined by the method adapted from Costa et al. (2006)23. The tumor necrosis factor alpha (TNF-α), interleukin 6 (IL-6) and interleukin 10 (IL-10), were measured by commercial immunoenzymatic assay (ELISA) (Fisher Scientific Term, Carlsbad, CA, USA).

Exercises Protocol

The exercises were based on the Guideline for the prescription of exercise to be given to people with HIV/AIDS24. The exercise session consisted of seven RE (chest press, leg press 45∘, lat pulldown machine, knee extension, triceps pulley, knee curl, and Scott biceps curl machine) involving different muscle groups with three sets per exercise.

The recovery interval adopted was of 90 seconds between the sets and 120 seconds between exercises. The number of repetitions used in each one of these sets was 8–12 repetitions, applying the fixed loads method25. The loads were compatible with the number of repetitions stipulated for each exercise. The load determination occurred during familiarization sessions.

Statistical Analysis

The descriptive statistics were used to characterize the sample. Shapiro-Wilk test was used for data distribution analysis. To compare inflammatory markers and oxidative stress, before and after exercise, analysis of variance of repeated measures (one-way ANOVA) followed by the Bonferroni post hoc test was used. The level of significance adopted for all analyzes was $P < 0.05$. The data were analyzed using SPSS Software (IBM, New York, United States). The power of the sample was calculated with a power of 80% and effect size = 0.05 (GPower software 3.19). Nine participants are required.

Results

The characteristics of the participants are presented in Table 1.

Table 1. Characterization of the participants of this study (n=10)

Variable	Mean ± Standard deviation	IC 95%
Age (years)	45.00 ± 12.77	35.86 – 54.14
Weight (kg)	65.72 ± 12.05	57.10 – 74.33
Height (m)	1.55 ± 0.05	1.50 – 1.58
BMI (kg/m²)	27.38 ± 4.01	24.51 – 30.25
CD4 (mm³)	388.70 ± 192.20	251.21 – 526.19
CD8 (mm³)	944.80 ± 539.59	558.80 – 1330.8
CD4/CD8	0.49 ± 0.31	0.27 – 0.71
Viral load (copies/ml)	77.20 ± 54.15	9.96 – 144.44
Diagnosis time (years)	10.37 ± 5.68	5.63 – 15.12
ART time (years)	7.75 ± 5.85	2.86 – 12.64

Notes: BMI = Body Mass Index; ART = Antiretroviral Therapy
Source: Authors

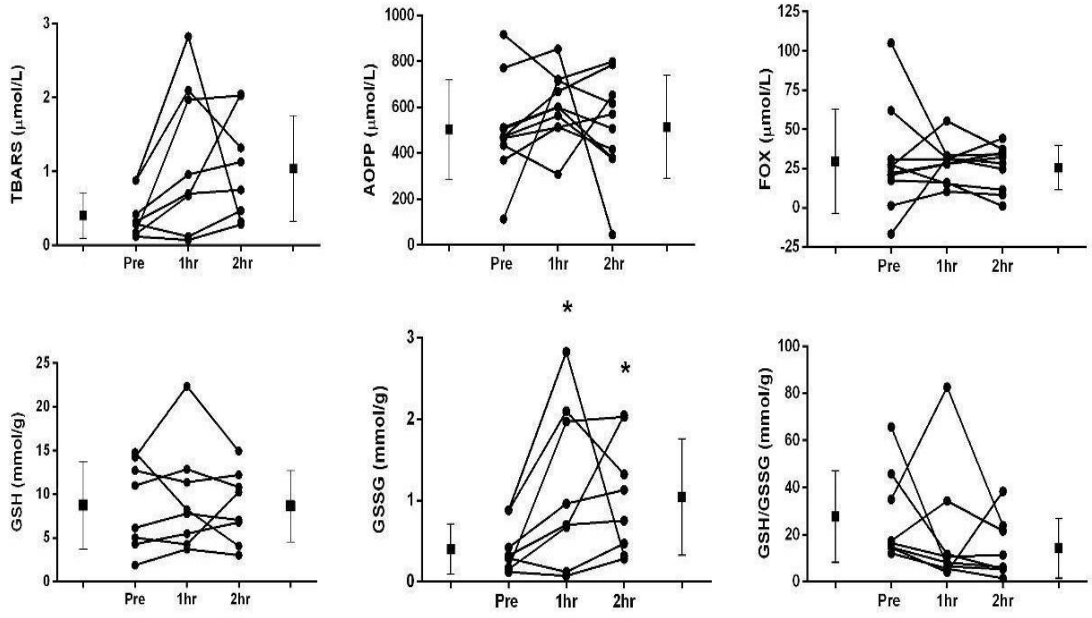

Figure 1. Effects of a single resistance exercise session on oxidative stress markers in women living with HIV.

Note: The circles represent individual data before (pre), after one hour (1h) and two hours (2h) of the exercise session. The squares represent the values of mean and standard deviation, before and after two hours of the session. TBARS = Thiobarbituric Acid Reactive Substances; AOPP = Advanced Oxidation Protein Products; FOX = Method of verification of Total Hydroperoxides; GSH = Reduced Glutathione; GSSG = Oxidized Glutathione; GSH / GSSG = Ratio between Reduced and Oxidized Glutathione. * $P \leq 0.05$ vs pre.

Source: Authors

Regarding the drugs used in ART, seven patients use a combination of Nucleoside Reverse Transcriptase Inhibitors (NRTIs) + Protease Inhibitors (PI); one used, NRTIs + Non-Nucleoside Reverse Transcriptase Inhibitors (NNRTIs); one, NRTIs + Integrase Inhibitors (II) and one NRTIs + PI + II.

Figure 1 shows the individual values of serum concentrations of oxidative stress markers at pre, 1hr, and 2hr after the RE session. For most markers, no differences were found, except for GSSG, where the values found over the dosages were (pre, post 1hr, and post 2hr) 0.40 ± 0.11 vs 1.18 ± 0.36 vs 1.04 ± 0.25 mmol/g hemoglobin, respectively. There was an increase over pre vs post 1hr ($p = 0.011$) and an increase over pre vs post 2hr ($p = 0.05$), representing a final mean increase of 160%.

The results of the inflammatory markers are shown in figure 2. For IL-6 the mean values were 2.47 ± 0.67 vs 3.63 ± 1.26 vs 5.38 ± 2.15 pg/ml, for the dosing times (pre, post 1hr, and post 2hr), with an increase of approximately 117% compared to pre vs post 2hr ($p = 0.014$), and 48% when compared to post 1hr vs post 2hr ($p = 0.017$). For TNF-α the mean values found over time (pre, post 1hr, and post 2hr) were 4.60 ± 0.55 vs 6.95 ± 0, 77 vs 9.10 ± 1.03 pg/ml, respectively, with a difference between pre vs post 1hr ($p = 0.047$), with an increase of 51%, and difference between pre vs post 2hr ($p = 0.029$) representing an increase of 98%. Finally, the mean IL-10 values were 3.34 ± 1.25 vs 3.32 ± 0.64 vs 2.73 ± 0.66 pg/ml, pre, post 1hr, and post 2hr, respectively, without statistical difference between time points.

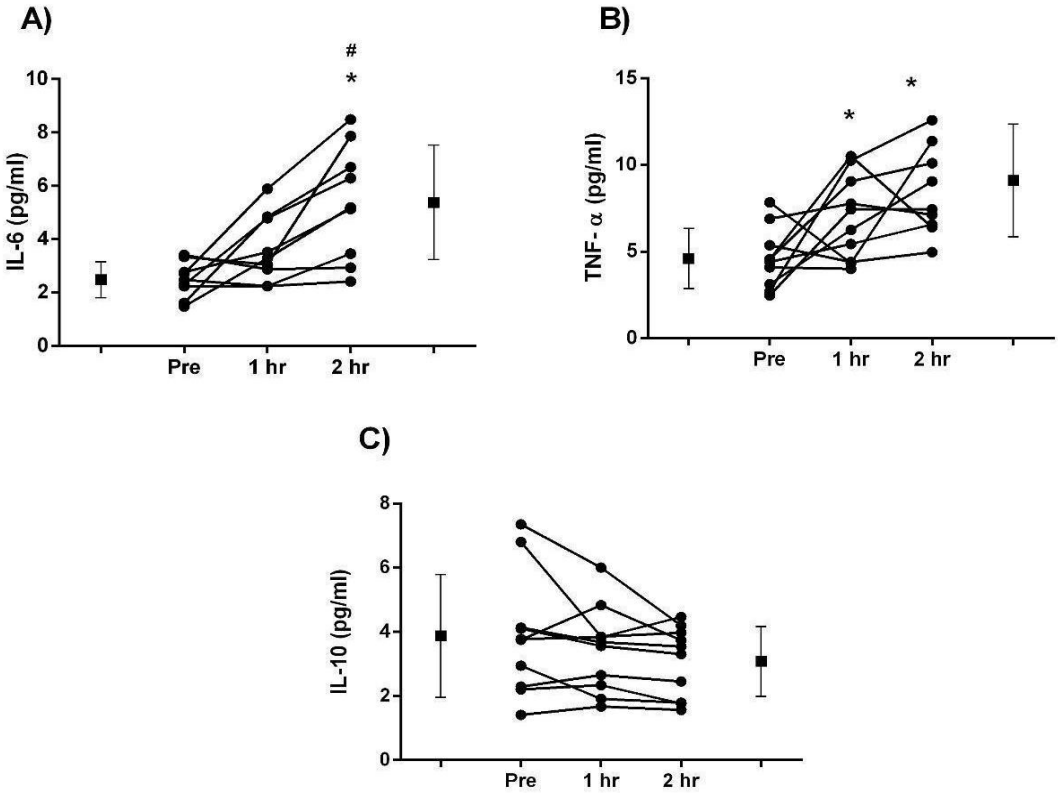

Figure 2. Serum concentrations of inflammatory markers, IL-6 (panel A), TNF-α (panel B) and IL-10 (panel C) pre, post 1 hr and post 2hr of a single resistance exercise session.

Note: The circles represent individual data before (pre), after one hour (1hr), and two hours (2hr) of the exercise session. The squares represent the values of mean and standard deviation, before and after two hours of the session. TNF-α = Alpha Tumor Necrosis Factor; IL-6 = Interleukin 6; IL-10 = Interleukin 10. * $P \leq 0.05$ vs pre. #$P \leq 0.05$ vs 1hr.

Source: Authors

Discussion

The results show that an RE indicated high levels of GSSG, but it is noteworthy that there were no changes in the levels of GSH, nor in the GSH/GSSG ratio, which would be important to prove an increase in oxidative stress. Furthermore, the levels of TNF-α and IL-6 cytokines, in general, significantly increased after 2 hours of the session. IL-10, on the other hand, showed no statistical difference. These findings partly confirm the initial hypothesis that acute exercise could cause an inflammatory reaction and oxidative stress in PLH.

Exercise practiced at moderate intensity in PLH could harm these patients, which would probably not happen with a healthy individual, since the guidelines recommend such intensity[26,27]. However, submitting any individual with or without HIV and not accustomed to a RE protocol, which has eccentric actions, would run the risk of triggering an acute physiological stress[16,28], capable of generating a natural response of the organism involving inflammation[29] and oxidative stress[30]. This response was not yet known in PLH. The intensity proposed in the study is the minimum load necessary for the body, after several exposures to the aggressor, to induce adaptive and beneficial mechanisms, where the literature has addressed possible protective effects of exercise against inflammation[31] and oxidative stress[17].

The increase found in the oxidative and inflammatory stress marker was expected. However, it was not exacerbated, which makes RE of moderate intensity safe and tolerated by PLH. Continuously, what we must observe is whether these alterations intensify, otherwise there will be an overproduction of reactive oxygen species[32] and pro-inflammatory cytokines[33-35], which can worsen the situation of PLH, once they already have inflammation caused by the virus and use of ART[36,37].

It is worth noting that the sample was composed only of female PLH, eliminating possible interference in the results of gender differences, since, according to Fragala et al., (2011)[38] men and women differ in relation to the immune and oxidative systems. This aspect is a strength of our research as most studies related to this topic have been conducted with males. In addition, we highlight the need for further investigations with a sample composed of women, and also studies with a chronic design, to understand the actual behavior of these markers in PLH. As for limitations, there was the absence of a control group and the limited sample size. However, this is a factor identified as a difficulty in working with the population in question, especially when it comes to interventions with physical exercise[39].

The protocol of a single RE session in the present study offers women with HIV a simple and accessible modality that can be performed in gyms. Despite the expected increase in inflammatory markers and oxidative stress, these increases were not exacerbated. This indicates that a single bout of RE appears to be safe for women with HIV. As this is a study in which patients were supervised during the training session, we would like to suggest that this session be carried out under the supervision of a qualified trainer for better use of the protocol. Furthermore, it is reasonable to suggest that the current protocol could be incorporated into an inpatient or outpatient program to care for these patients.

Conclusion

The results of the present study indicated that one resistance exercise session increased the levels of inflammatory markers and oxidative stress in PLH in an expected and non-exacerbated manner, it is possible to conclude that resistance exercise at moderate intensity is safe for this population.

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