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Original article

Acute effects of high- and low-intensity exercise bouts on leukocyte counts

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Abstract

Background/Objective: It is widely accepted that physical exercise may bring about changes in the immune system. Even acute bouts of exercise can alter the number and function of leukocytes, but the degree of white blood cell trafficking depends on the intensity and duration of exercise. The aim of this study was to analyze the acute and short-term effects of exercise intensity on leukocyte counts and leukocyte subsets.
Methods: Nine physically healthy, active young males (21.0 ± 1.9 years) underwent three experimental trials: high exercise intensity [80% peak oxygen consumption (VO_{2peak})], low exercise intensity (40% VO_{2peak}), and the control condition (no exercise). Blood samples were collected prior to exercise, immediately after exercise, and 2 hours after exercise. Two-way analysis of variance for repeated measures was used to evaluate differences between the trials and the time-points, and to compare times within trials.
Results: There was a greater increase in the leukocyte count after high-intensity exercise, compared to the control condition ($p < 0.01$) and low-intensity exercise ($p < 0.01$). This effect was still present 2 hours after passive recovery ($p < 0.01$).
Conclusion: When the same participants were submitted to different exercise intensities, the acute and short-term effects of exercise on white blood cells were intensity-dependent immediately after exercise (i.e., lymphocytosis and monocytosis) and 2 hours after passive recovery (i.e., neutrophilia).

Keywords: Aerobic exercise; Immune system; Inflammatory response; White blood cell

Introduction

In clinical practice, total leukocyte counts and subsets are widely used to confirm inflammatory process-related acute immune system disturbances[1] that have been associated with the development of a number of conditions harmful to health.[2] However, a complex interplay between manifold genetic and environmental factors determine interpersonal variability in leukocytes.[3] A high interpersonal variation in white blood cell (WBC) counts has also been reported in physically active individuals.[4,5]

Despite the high interpersonal variation, it is widely accepted that physical exercise may promote changes in the immune system.[6,7] Even acute bouts of exercise can alter the number and function of leukocytes.[8] The degree of WBC trafficking depends on the intensity and duration of exercise.[9] It has been postulated

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that exercise increases stress-induced changes in the immune-neuroendocrine axis and in the circulating levels of metabolites that directly influence the function of immune cells.[10]

Natale et al[11] previously reported that aerobic training and resistance training induce changes in leukocyte counts in moderately fit individuals; by contrast, Nieman et al[12] did not find continuous or intermittent cycling on WBC in well-trained cyclists. Kendall et al[13] showed that the effects of exercise on leukocytes depend on the intensity, duration, and fitness level of individuals. These contradictory observations are intriguing. A variety of factors may have contributed to the conflicting data such as the fitness status of the individuals and differences in the exercise type, intensity, and duration. It is noteworthy that none of the previous studies evaluated the effects of different exercise intensities on leukocyte counts and subsets by standardizing the energy expenditure of the sessions and controlling the interpersonal variation in the WBC levels. The aim of this study was thus to analyze the acute and short-term effects of a single bout of aerobic exercise at low intensity and at high intensity on leukocyte counts and subsets in physically active young adults.

Methods

Participants

Nine healthy physically active young men (21.11 ± 1.90 years) of normal weight [body mass index (BMI), 19.9–25 kg/m^2] volunteered for this study. The exclusion criteria were chronic alcohol consumption, smoking and/or use of nutritional supplements, metabolic or endocrine diseases, and use of any anti-inflammatory drug. The study was conducted in accordance with the Helsinki Declaration and was approved by the University of Pernambuco's Ethics Committee (Recife, Brazil; approval number 154/08). All volunteers gave written, informed consent.

Experimental design

In this crossover study, the participants were assigned by simple randomization to three experimental sessions with a 7-day wash out: (1) the control session in which the participants remained seated for 2 hours (i.e., no exercise); (2) the low-intensity exercise (LIE) session in which the participants exercised on a treadmill at an intensity corresponding to 40% of peak oxygen consumption (VO_{2peak}); and (3) the high-intensity exercise (HIE) session in which the participants exercised on a treadmill at 80% VO_{2peak}. For the LIE and HIE sessions, the energy expenditure was set at 350 kcal, estimated by indirect calorimetry (e.g., direct gas analyses), based on the metabolic equivalent of task (MET). The energy expenditure is 4.96 kcal for each liter of O_2 consumed. All sessions were conducted under a steady-state condition and with a respiratory exchange ratio < 1.0 (control, 0.82 ± 0.05; LIE, 0.90 ± 0.04; HIE, 0.94 ± 0.05). Each participant served as his own control.

During the first visit to the laboratory, anthropometry and body composition were measured and VO_{2peak} was determined. On the remaining visits, the participants arrived at the laboratory around 7:00 AM, after fasting overnight. They were then weighed and were served a standard snack (350 kcal composed of 61.7% carbohydrates, 13.44% proteins, and 24.86% lipids). At 7:30 AM, the participants underwent a LIE session, a HIE session, or the control session, and subsequently remained seated for 2 hours. The trials were conducted in a temperature-controlled room (21–23°C) in randomized and were conducted at the same time of the day to avoid any circadian variations. Participants were asked to refrain from vigorous exercise for 48 hours prior to the sessions.

Anthropometry and body composition

The participants were weighed on a Filizola scale (Model 160/300; São Paulo, Brazil) to the nearest 0.1 kg while wearing light clothing and no shoes. Height was measured to the nearest 0.5 cm using a wall-mounted Filizola stadiometer (Model 160/300; São Paulo, Brazil). Body composition was determined by bioelectrical impedance (Biodynamics A-310 body composition analyzer; Biodynamics Corporation, Shoreline, New York, USA).[12]

VO_{2peak} determination

Oxygen consumption (VO_2) was directly measured using a continuous incremental treadmill test (Super ATL; Inbrasport, Porto Alegre, Brazil), as previously described. The inclination was set at 1.0%, and the initial workload was 5.0 km/h (4 minutes). The speed was thereafter increased to 1.0 km/h every minute. The termination criteria were volitional fatigue, a Borg scale value > 18, and gas exchange ratio > 1.15. The greatest VO_2 obtained prior to the test interruption was the VO_{2peak}. The VO_2 and carbon dioxide production (VCO_2) were analyzed breath by breath and displayed every 15 seconds in an open circuit respiratory metabolic system (Metalyzer IIB; Cortex Biophysik, Leipzig, Germany).

Blood leukocyte and subset counts

The complete routine tests for WBCs was performed using an automated method (Abbott Cell-Dyn 3700 Hematology Analyzer Features; Abbott Laboratories, St. Ana, USA). For the exercise trials, blood samples were obtained prior to the onset of exercise (i.e., baseline), immediately after exercise (i.e., acute), and 120 minutes after exercise (i.e., short-term). For the control trial, samples were collected at baseline, after 30 minutes (i.e., acute), and 120 minutes after baseline (i.e., short-term). The WBC count procedures were blindly performed.

Statistical analysis

All data were expressed by the mean and standard deviation. Analysis was performed using Statistica 8.0 software (Statistica 8, 2008; Statsoft Inc., Tulsa, OK, USA). Two-way analysis of variance (ANOVA) for repeated measures was used to evaluate differences between trials (i.e., control, LIE,

and HIE) and time-points (i.e., baseline, acute and short-term). One-way ANOVA was issued for comparisons within trials using the Tukey test as the *post hoc* test in all trials. Statistical significance was set at $p < 0.05$. With an alpha of 0.05 and power of 0.80, nine individuals for each condition was sufficient to detect significant differences > 2188 counts.

Results

Eleven of 20 participants did not complete all experimental sessions (2 participants did not complete the sessions because of health problems and 3 participants did not complete all experimental procedures); thus, nine individuals were included in the analysis. The mean age, body mass, BMI, fat mass, and VO_{2peak} of the volunteers were 21.11 ± 1.19 years, 65.39 ± 7.21 kg, 22.06 ± 1.36 kg/m^2, 13.71% ± 2.94%, and 49.77 ± 5.71 mL/(kg × min), respectively. The speed was lower (5.6 ± 0.34 km/h) during LIE trial than during the HIE trial (11.28 ± 0.77 km/h) ($p < 0.001$). However, the LIE lasted longer (59.68 ± 5.02 minutes) than HIE (29.43 ± 2.52 minutes; $p < 0.001$) with no difference between conditions in energy expenditure (LIE was 339.35 ± 20.16 kcal and HIE was 341.75 ± 17.62 kcal; $p > 0.05$).

The total leukocyte count did not differ from trial to trial at baseline; however, there was a major acute effect in HIE, compared to the control trial ($p < 0.01$) and LIE ($p < 0.01$). Two hours after exercise, HIE-induced leukocytosis could still be observed ($p < 0.01$). Neither LIE ($p = 0.98$) nor time ($p = 0.98$) had any effect (Fig. 1).

At the baseline, no differences existed from one trial to another for monocytes, lymphocytes, or neutrophils. The LIE session did not modify the leukocyte subsets at any time point ($p > 0.05$), whereas the HIE produced transitory acute lymphocytosis ($p < 0.01$; Fig. 2) and monocytosis ($p < 0.01$; Fig. 3), which returned to normal 2 hours after the physical exertion ended. The data do not show an acute effect by HIE on neutrophils, although there was an increase in neutrophils after 2 hours ($p < 0.01$; Fig. 4).

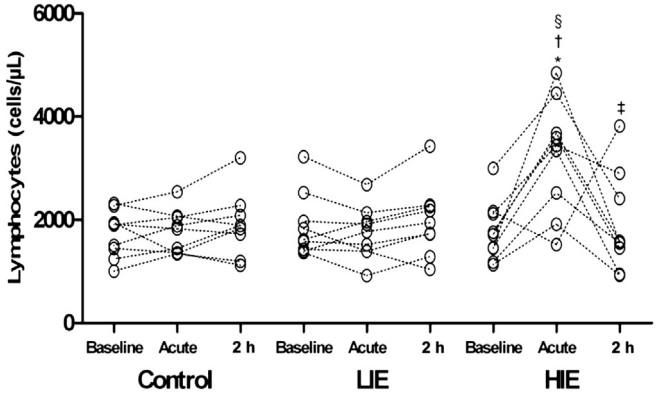

Fig. 2. Lymphocyte concentration in response to different exercise intensities *versus the baseline, ‡versus the acute session, †versus the control session, and §versus LIE. HIE = high-intensity exercise; LIE = low-intensity exercise.

Discussion

The main findings of this study were: (1) HIE causes significant leukocytosis, regardless of interindividual variability; (2) leukocytosis is mediated by transitory lymphocytosis and monocytosis, followed by neutrophilia; and (3) LIE is incapable of modulating the immune system. Acute exposure to short-term stress (e.g., physical exercise) can lead to changes in WBC.[9,14] Similar to our results, previous studies have reported alterations in leukocyte and subset counts in response to HIE.[15–18]

Acute exercise mobilizes leukocytes. This leads to leukocytosis mediated by an increased number of circulating and tissue lymphocytes.[19] These alterations are dependent on the intensity and duration of the exertion.[7] It has been suggested that the brain releases neuroendocrine mediators that help the immune system, once an organism is stressed.[20] Exercise bouts are followed by an intensity-dependent increase in sympathetic activity (e.g., catecholamine release).[21] Thus, the greater the intensity of exercise, the greater the increase in the circulating levels of epinephrine and norepinephrine (NOR) with subsequent mobilization of white immune cells.[11]

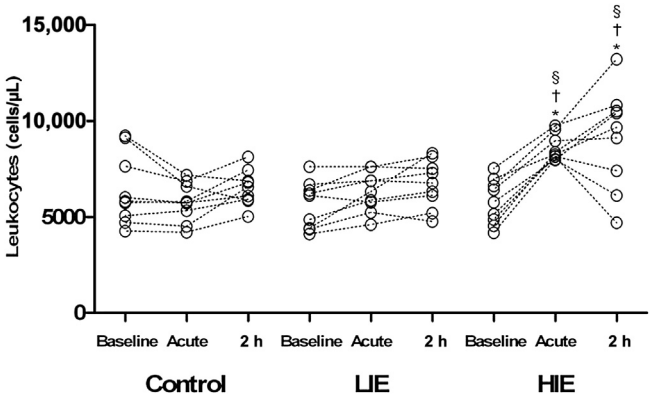

Fig. 1. Leukocyte concentration in response to different exercise intensities in physically active men *versus the baseline, †versus the control session, and §versus LIE. HIE = high-intensity exercise; LIE = low-intensity exercise.

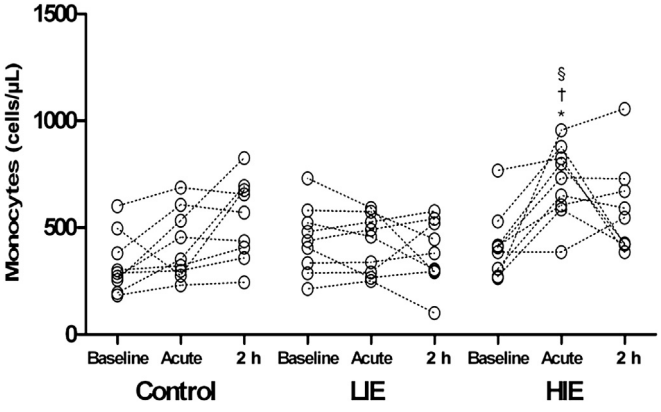

Fig. 3. Monocyte concentration in response to different exercise intensities *versus the baseline, †versus the control session, and §versus the LIE. HIE = high-intensity exercise; LIE = low-intensity exercise.

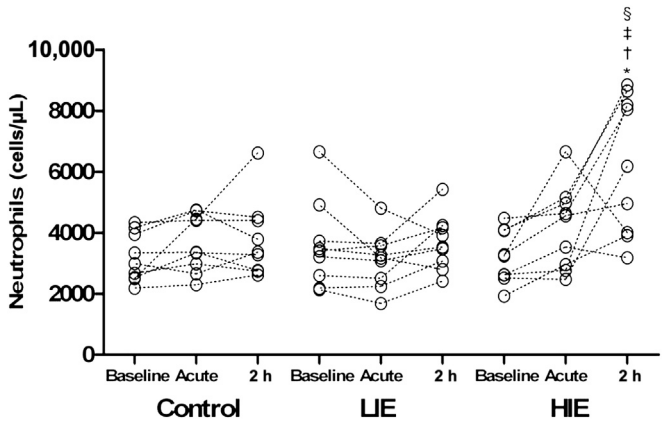

Fig. 4. Neutrophil concentration in response to different exercise intensities *versus the baseline, ‡versus acute exercise, †versus the control session, and §versus LIE. HIE = high-intensity exercise; LIE = low-intensity exercise.

Our results demonstrated that the acute effect of HIE on leukocyte counts is lymphocyte- and monocyte-dependent. This increase in lymphocytes could be explained, at least partially, by the fact that lymphoid organs are innervated by sympathetic nerve fibers that respond to stimuli by releasing epinephrine and NOR.[19] In the blood, epinephrine and NOR recruit lymphocytes[7] and monocytes[22] because these cells have a high density of β-receptors. The strongest point of the present study is the fact that it is the first to analyze the effects of exercise intensity on WBC in humans, and it minimized bias by subjecting the same individuals to all experimental conditions so that they acted as their own control.

The increase in lymphocytes and monocytes is transient and the levels of these cells return to normal after 2 hours.[11,12] There is a significant rise in the lymphocyte apoptosis ratio when the exercise intensity is increased,[6] and elevated apoptosis in monocytes and lymphocytes after exercise (85% VO_{2max}).[23]

Athletes who undergo different intensities of physical exertion exhibit changes in the mitochondria of leukocytes and intensive exercise causes DNA damage in lymphocytes.[24,25] Leukocyte apoptosis also has an important role in maintaining lymphoid tissue homeostasis and avoiding immune-activation.[8,26] Some evidence suggests that glutamine metabolism delays spontaneous apoptosis in neutrophils in humans and rats.[27] During intense exercise, the spleen, thymus, and lymph nodes are exposed to glutamine deficiency because the demand from other tissues for glutamine is higher than the quantity produced by skeletal muscle; this affects the number and the function of leukocytes.[28,29]

Our study showed that persistent leukocytosis was neutrophil-dependent, a fact confirmed in other findings.[7,30] Lymphocytes and monocytes show an early increase; by contrast, the recruitment of neutrophils is independent of adrenergic mechanisms.[31] The delayed increase in neutrophils may be mediated by blood cortisol[32] and/or by exercise-related tissue damage or by an increase in chemokines.[33]

The present study has some limitations that should be considered. We were unable to assess the circulating levels of cytokines and activity of the cells. Future studies that include these factors in a larger population may shed light on the underlying mechanisms and allow generalization of these findings.

In short, HIE is capable of altering the count of the leukocytes and their subsets (i.e., neutrophil, monocytes, and lymphocytes) immediately after exercise and after 2 hours without any interpersonal variation. These changes suggest that the short-term effects of exercise on the immune system are intensity-dependent. We thus concluded that a single bout of high-intensity aerobic exercise is able to create disturbances in the immune system. Such information should be used to improve the quality of exercise prescription, and support physicians in better managing susceptibility to upper respiratory tract infections in athletes and people with chronic low-grade inflammation when exercising.

Based on current knowledge regarding exercise and WBCs, there is an association between exercise and improvements in health such as a reduced risk of cardiovascular disease,[16] diabetes and impaired fast glucose,[34] and atherosclerosis,[35] and increased immunocompetence.[9] Further studies should therefore be conducted to shed more light on the effects on the immune system by long-term exposure to physical exercise at different intensities in different types of individuals such as athletes, sedentary individuals, patients with chronic diseases, older people, and children.

Conflicts of interest

The authors have no financial or other conflicts of interest to declare.

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