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Article

Effects of Two Different Self-Paced Training Modalities on the Aerobic Fitness Level, Psychophysiological Responses and Antioxidant Status in Young Well-Trained Adults

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Abstract: This study aims to investigate the effects of self-paced high-intensity interval training (Sp-HIIT) vs self-paced moderate-intensity continuous training (Sp-MICT) on the aerobic fitness level, psychophysiological responses and antioxidant status, to assess the relationship between aerobic fitness level and antioxidant markers. Well-trained males were randomised into Sp-HIIT and Sp-MICT. The intervention consisted of 3 weekly sessions during an 8-wk period. Sp-HIIT performed two sets of 12–24 x 30-s high-intensity runs ≥85 %HR_{max} followed by 30-s rest periods, while Sp-MICT performed 24-48 min of continuous running at 60-75 %HR_{max}. Pre and post-intervention testing included maximal oxygen uptake (VO_{2max}) assessment during a 30-15 intermittent fitness test (30-15 IFT), as well as resting blood samples analysed for oxidative stress markers (Malondialdehyde (MDA)) and intracellular antioxidant enzymes activity (Catalase (CAT), Superoxide Dismutase (SOD) and Reduced (GSH) and Oxidized Glutathione (GSSG)). The Sp-HIIT showed meaningfully greater improvement in velocity of 30-15 IFT, VO_{2max} and MDA responses. Furthermore, the Sp-HIIT demonstrated higher psychophysiological responses than the Sp-MICT, except for anger responses. In conclusion, these results suggest that Sp-HIIT has higher level of exercise-induced beneficial effects in physiological responses with greater perceived exertion in young well-trained adults.

Keywords: Interval training; continuous training; mood; oxidative stress; perceived exertion

1. Introduction

It is widely known that physical inactivity associates with numerous chronic diseases [1] which also has led to the recommendation from the World Health Organization that all adults should perform regular physical activity [2]. The positive influence of systematic exercise has a multicomponent impact on human health including decreases in coronary heart diseases risk [3] and obesity [4], supports type 2 diabetes treatment [5], and improves sleep quality [3] as well as mental health [6]. However, the efficiency of different types of physical activity could vary depending on volume and exercise intensity. Therefore, scientific verification of applied training programs seems to be an important task for researchers.

The modern lifestyle allows people to devote limited time to physical activity. Thus, since the beginning of the 21st century, the topic of low-volume, but high-intensity interval training has drawn attention and explored extensively [7,8]. It appears that repeated high-intensity training could be time-efficient exercise mode to induce positive adaptations skeletal muscle metabolism and to upregulate the aerobic capacity in men and women [9,10]. Weston et al. [11] introduced definitions of the most popular strategies of interval training based on exercise intensity. High-intensity interval training (HIIT) includes efforts at intensity exceeding 80-85% of maximal heart rate, while sprint interval training (SIT) is defined as efforts at an intensity equal to or greater (supramaximal) than peak aerobic capacity. Moreover, to describe the continuous efforts performed at intensities lower than HIIT, moderate-intensity continuous training (MICT) is widely used [12].

The effectiveness of mentioned above training forms has often been compared. The first such comparisons, published nearly half a decade ago [13], reported no differences in maximal oxygen uptake (VO_{2max}) changes and physiological responses between continuous and interval training participants. However, more recent studies demonstrated that interval training could provide even more benefits in physiological adaptations when training volume is matched [9]. Even when interval training volume is lower, the adaptive response has been shown to be comparable or even superior compared to endurance training [10,14,15]. Ramos et al. [16] found that HIIT enhances vascular function more effectively compared to MICT. Furthermore, Milanović et al. [17] observed more significant improvements in VO_{2max} following HIIT than after traditional endurance training, which is supported by Nybo et al [15]. Similar tendencies were noted in psychological responses. It was well established that both HIIT and MICT are beneficial in improving mental health [18]. Previous research indicated that after HIIT, participants experienced equal or greater post-exercise enjoyment than after MICT [19,20]. However, repeated intense exercise results in increased reactive oxygen species (ROS) generation, which is associated with cellular function disruption [21]. Nevertheless, this negative effect could be reduced by antioxidant defence systems, including Catalase (CAT), Superoxide Dismutase (SOD) and Reduced (GSH) and Oxidized Glutathione (GSSG) [22]. Although the acute effect of HIIT efforts on the antioxidant status has been accurately described [23], the number of studies investigating the influence of training programs in humans is still sparse. Complete evaluation of the training effectiveness usually covers three main areas: changes in physical performance, a physiological response and psychological reaction. All these elements should be considered when assessing the training plan's efficiency. Therefore, the purpose of this research was twofold: (1) to evaluate the effects of self-paced high-intensity interval training (Sp-HIIT) vs self-paced moderate-intensity continuous training (Sp-MICT) on aerobic fitness level, psychophysiological responses and antioxidant status in young adults; (2) to identify potential correlations between the aerobic fitness level and oxidative stress and antioxidant markers.

2. Materials and Methods

Experimental approach to the problem

The effects of Sp-HIIT vs Sp-MICT on young adults' aerobic fitness level, psychophysiological reactions, and antioxidant status were compared using a two-group, parallel research design. Pre and post-intervention measurements included a skinfold measurement, 30-15 Intermittent Fitness Test (30–15 IFT), serum oxidative stress marker [Malondialdehyde (MDA)] and antioxidant markers [Catalase (CAT), Superoxide Dismutase (SOD), Reduced (GSH) and Oxidized Glutathione (GSSG)]. Twenty-four randomized recreationally male young adults performed either the Sp-HIIT or the Sp-MICT group. They were randomly assigned to one of the groups. Two 12–24 x 30-second high-intensity runs ≥85 $\%HR_{max}$ were completed by the Sp-HIIT group, followed by 30-second rest periods. The participants in Sp-MICT performed 24-48 minutes of continuous running at 60-75 $\%HR_{max}$. These eight-week training interventions were performed three times a week, and two-day intervals separated each training session. All measurements were performed on standard outdoor athletics track with a tartan surface between 3 and 6 o'clock in the afternoon for similar chronobiological responses. They were told to keep a regular nutritional intake both before and during the trial, and they were familiar with testing and training techniques.

Design

Before the study began, we estimated the sample size using the G*Power software program version 3.1 (Düsseldorf, Germany). After adding a partial effect size of 0.2, a power of 0.8, a p-value of 0.5 (2 groups and 2 number of measurements), and a correlation of 0.5, considering previous study findings of imposed vs self-selected training [24]. We found a recommended total sample size of 12 for the study.

Participants

Twenty-four young male adults were randomly assigned to a Sp-HIIT (n = 12; age: 21.8 ± 1.3 years, height: 176.6 ± 5.8 cm, weight: 76.9 ± 10.6 kg) or the Sp-MICT (n = 12; age: 22.1 ± 1.6 years, height: 179.5 ± 5.9 cm, weight: 78.2 ± 12.2 kg). All participants were involved in team sports including handball, basketball, and soccer, had at least two years of experience with the training workload of four training units per week, which included core strength training, aerobic activity, and group exercise. Before the study began, written informed permission was acquired from the participants after they had been told of the research's requirements, benefits, risks, and procedures.

Procedures

Biochemical Procedure. A minimum of 10 hours fasting before their routine breakfast time (7:00-8:30 am), blood samples from each participant were collected into lithium heparin vacutainers and centrifuged at 5000 rpm for 10 min at four °C to separate plasma. The plasma samples were collected and frozen at -80°C until biochemical analysis. The spectrophotometric method was used to determine CAT [25], SOD [26] activities and MDA levels [27]. Furthermore, GSH and GSSG concentrations were also determined spectrophotometrically, as described in a previous study [28]. After the blood collection to analyse oxidative stress with MDA and antioxidant markers, the body fat percentages of participants, using the Holtain Tanner–Whitehouse skinfold calliper (Holtain, UK) and Gulick anthropometric tape (Holtain), were estimated using the validated formula for Turkish athlete [29]. Following the anthropometric measurements, each participant performed the 30–15 IFT. This test, consisting of 30 s of running and 15 s of passive recovery, is an acoustically and reliably progressive test according to the procedures described. After the 30–15 IFT, the maximal oxygen uptake (VO_{2max}) was estimated from the maximum speed (30-15 V_{IFT}) reached in the last stage of the test [30].

Table 1. 8 weeks training programmes and the weekly rating of perceived exertion and mood responses.

	Sp-MICT (*n* = 12)						Sp-HIIT (*n* = 12)					
Weeks	Training	RPE	Fatigue	Depression	Anger	Vigour	Training	RPE	Fatigue	Depression	Anger	Vigour
1	24 min cont.	12.5 ± 0.3	2.4 ± 0.4	1.7 ± 1.0	1.6 ± 0.8	1.4 ± 0.4*	2 x (12 x 30 s),	16.2 ± 0.2*	3.0 ± 0.4	3.2 ± 1.3*	1.6 ± 1.0	0.9 ± 0.4
2	running	13.2 ± 0.3	2.9 ± 0.5	1.6 ± 0.5	1.6 ± 0.2	1.1 ± 0.4*	30 s rest	17.1 ± 0.1*	3.3 ± 0.4	2.3 ± 0.9*	1.3 ± 0.4	0.7 ± 0.4
3	32 min cont.	13.1 ± 0.5	3.0 ± 0.4	1.6 ± 0.5	1.3 ± 0.2	0.9 ± 0.4*	2 x (16 x 30 s),	17.1 ± 0.2*	3.3 ± 0.3	2.0 ± 0.6*	1.3 ± 0.3	0.6 ± 0.3
4	running	14.1 ± 0.4	2.9 ± 0.6	1.1 ± 0.6	1.1 ± 0.4	1.1 ± 0.6*	30 s rest	17.3 ± 0.1*	3.2 ± 0.4	2.1 ± 0.6*	1.1 ± 0.5	1.8 ± 0.4
5	40 min cont.	13.6 ± 0.7	2.8 ± 0.4	1.4 ± 0.6	1.1 ± 0.5	1.2 ± 0.4*	2 x (20 x 30 s),	17.4 ± 0.1*	3.1 ± 0.3	2.2 ± 0.6*	0.9 ± 0.4	0.9 ± 0.3
6	running	13.4 ± 0.1	2.8 ± 0.5	0.9 ± 0.5	1.1 ± 0.4	0.9 ± 0.4*	30 s rest	17.8 ± 0.4*	3.2 ± 0.3	1.3 ± 0.4*	0.9 ± 0.3	0.7 ± 0.3
7	48 min cont.	13.6 ± 0.3	3.9 ± 0.5	0.8 ± 0.4	1.1 ± 0.5	1.1 ± 0.5*	2 x (24 x 30 s),	17.7 ± 0.3*	3.3 ± 0.3	0.9 ± 0.5*	0.8 ± 0.4	0.7 ± 0.3
8	running	13.7 ± 0.6	2.9 ± 0.6	0.8 ± 0.7	0.9 ± 0.6	1.0 ± 0.5*	30 s rest	17.9 ± 0.3*	3.4 ± 0.3	1.0 ± 0.6*	0.8 ± 0.4	0.6 ± 0.3

Sp-HIIT: high-intensity interval training; Sp-MICT: moderate-intensity continuous training; RPE: rating of perceived.

Training Interventions. Sp-HIIT and Sp-MICT interventions were performed on standard athletics track 3 days per week for eight weeks, and two-day intervals separated each training session to maximize physical and physiological performance. A progressive training design was developed to increase final performance in both training programmes. The Sp-MICT group's total training duration was divided in half during the research in accordance with the Sp-HIIT group. Two 12–24 x 30-second high-intensity runs ≥85 $\%HR_{max}$ were completed by the Sp-HIIT group, followed by 30-second passive resting. The participants in Sp-MICT continuously performed 24-48 minutes of running at 60-75 $\%HR_{max}$. Detailed information about training is summarized in Table 1. A 15-minute standardized warm-up, which included 10 minutes of running and 5 minutes of static and dynamic stretching activities, was the first part of each training session. Our participants were allowed to change and maintain their pace during each training session without receiving any guidance on the intensity of their activity. The rating of perceived exertion (RPE) was determined using the category level (CR-20) Borg scale immediately after each session [31]. Brunel Mood Scale (BRUMS) was used to determine their mood profile [32,33] before and after the training sessions. The validated and reliable this scale, consists of 19 items and four sub-scales (e.g., fatigue, depression, anger and vigour) scored on a five-point Likert scale [34]. Using the responses to a question of "how do you feel right now?" volunteers indicated whether they experienced such feelings on a 5-point scale (0 = not at all; 1 = a little; 2 = moderately; 3 = quite a bit; 4 = extremely).

Statistical analysis

Data were represented as mean ± SD. A mixed ANOVA was used to test for interactions and the main effects of time and group on aerobic fitness level, psychophysiological responses and antioxidant status. Correlation between aerobic fitness level and pro/antioxidant markers was performed using the Pearson (r) or Spearman (rho) correlation coefficient. The correlations were considered trivial (<0.1), small (0.1 to <0.3), moderate (0.3 to <0.5), large (0.5 to <0.7) and very large (0.7 to <0.9), extremely large (0.9 – 1.0) [35]. SPSS version 24.0 was used to conduct all statistical analyses (SPSS, Version 24.0 for Windows; SPSS Inc., Chicago, IL, United States). Statistical significance was set at the level of $p \leq 0.05$.

3. Results

The weekly RPE and mood responses are demonstrated in Table 1. Overall, during the 8-week self-paced training period, the Sp-HIIT showed higher RPE responses than those to Sp-MICT sessions ($p \leq 0.05$, d = ranging from 7.6 to 17.4). Moreover, the Sp-HIIT also showed higher depression responses than those to Sp-MICT sessions ($p \leq 0.05$, d = ranging from 0.2 to 4.1). In contrast, the vigour responses from the Sp-MICT were significantly higher than those to Sp-HIIT sessions ($p \leq 0.05$, d = ranging from 0.2 to 1.3).

Table 2. Effect of 8 weeks trainings on anthropometric, aerobic fitness level and antioxidant status of the participants.

	Sp-MICT (*n* = 12)				Sp-HIIT (*n* = 12)			
	Pre-test	Post-test	Cohen's *d*	Descriptor	Pre-test	Post-test	Cohen's *d*	Descriptor
Body weight (kg)	78.2±12.2	76.1±11.8*	0.2	trivial	75.7±8.9	73.2±8.6*	0.3	small
Body fat (%)	14.8±3.7	13.2±3.2*	0.4	small	13.9 ±3.0	11.8±2.4*	0.8	moderate
BMI ($kg.m^{-2}$)	24.2±2.9	23.5±2.8*	0.2	small	25.0±2.4	24.2±2.3*	0.3	small
30-15 V_{IFT} ($km.h^{-1}$)	14.5±0.5	15.1±0.6*	1.0	moderate	14.3±0.6	16.4±0.7*♯	3.2	very large
VO_{2max} ($ml.min^{-1}.kg^{-1}$)	40.7±1.3	42.1±1.5*	1.1	moderate	40.2±1.5	45.1±1.6*♯	3.1	very large
MDA ($mmol.ml^{-1}$)	0.4±0.0	0.4±0.0*	2.0	very large	0.4±0.0	0.5±0.0*♯	3.2	very large
CAT ($U.mg^{-1}$ Hb)	66.9±11.1	52.0±11.5*	1.3	large	64.5±6.3	44.8±4.5*	3.6	very large
SOD ($U.mg^{-1}$ Hb)	1.4±0.2	1.3±0.1*	0.9	moderate	1.4±0.2	1.2±0.2*	1.5	large
GSH ($\mu mol.g^{-1}$ Hb)	11.4±0.7	13.5±0.6*	3.3	very large	11.7±0.7	14.1±1.2*	2.4	very large
GSSG ($\mu mol.g^{-1}$ Hb)	8.9±1.0	10.5±1.1*	1.5	large	9.4±0.7	10.9±0.7*	2.1	very large
GSH/GSSG ratio	1.3±0.1	1.3±0.1*	0.1	trivial	1.2±0.1	1.3±0.2*	0.5	small

Data presented as mean ± SD. BMI: body mass index; 30-15 V_{IFT}: Maximum speed reached in the last stage of the 30-15 Intermittent Fitness Test; VO_{2max}: maximal oxygen uptake; CAT: catalase; SOD: superoxide dismutase; MDA: malondialdehyde; GSH: glutathione; GSSG: oxidized glutathione. * $p \leq 0.05$ for within-group changes. ♯ $p \leq 0.05$ for between-group changes.

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The Sp-HIIT and Sp-MICT interventions demonstrated similar anthropometric, performance responses, and antioxidant status improvements except for 30-15 VFIT, VO_{2max} and MDA responses in Table 2. The Sp-HIIT showed greater improvement in the 30-15 V_{IFT} (14.7%, $p \leq 0.05$, $d = 3.20$ [very large effect]), VO_{2max} (12.2%, $p \leq 0.05$, $d = 3.15$ [very large effect]) and MDA (11.2%, $p \leq 0.05$, $d = 3.59$ [very large effect]) compared with the Sp-MICT group.

Table 3. Correlations between aerobic fitness level **and** oxidative stress and antioxidant markers.

	CAT (U.mg^{-1} Hb)		SOD (U.mg^{-1} Hb)		MDA (mmol.ml^{-1})		GSH (µmol.g^{-1} Hb)		GSSG (µmol.g^{-1} Hb)		GSH/GSSG ratio	
	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post	Pre	Post
30-15 V_{IFT} (km.h^{-1})	-0.1 (small)	-0.1 (trivial)	-0.2 (small)	-0.2 (small)	-0.2 (small)	-0.1 (small)	-0.2 (small)	-0.3 (small)	-0.1 (small)	-0.3 (moderate)	0.1 (trivial)	0.2 (small)
VO_{2max} (ml.min^{-1}.kg^{-1})	0.0 (trivial)	0.0 (trivial)	0.2 (small)	0.1 (small)	0.1 (small)	0.1 (small)	0.1 (small)	0.2 (small)	0.1 (trivial)	0.4 (moderate)	-0.1 (trivial)	-0.3 (small)

Data presented as mean ± SD. 30-15 V_{IFT}: Maximum speed reached in the last stage of the 30-15 Intermittent Fitness Test; VO_{2max}: maximal oxygen uptake; CAT: catalase; SOD: superoxide dismutase; MDA: malondialdehyde; GSH: glutathione; GSSG: oxidized glutathione; * $p \leq 0.05$.

Table 3 demonstrates the correlations between aerobic fitness level, oxidative stress, and antioxidant markers during 8-week Sp-HIIT and Sp-MICT interventions. No significant differences were found in the oxidative stress and antioxidant markers with the aerobic fitness level between pre-and post-training.

4. Discussion

The main findings in our study were that both the Sp-HIIT and the Sp-MICT intervention improved body composition, aerobic fitness, and circulatory antioxidant status. However, the Sp-HIIT intervention caused greater improvement in the 30-15 V_{IFT}, VO_{2max}, and MDA responses, but with higher physical exertion, vigour and depression scores in comparison to Sp-MICT. Furthermore, no significant training-induced changes were found in serum oxidative stress and antioxidant markers.

The rating of psychophysiological responses, such as RPE-scores and mood profile, are validated, low-cost, and practical tools for measuring physical fatigue and psychological status in athletes. Our results are consistent with previous studies [13,36]. Thus, the HIIT-intervention induced higher negative changes in mood profile compared to the MICT. In addition to these supporting findings, the physical tiredness after a training session induced increased adverse feelings such as anxiety, anger, and depression [37,38]. The nature of HIIT and MICT might explain differences in mood state responses. While HIIT included running intervals of different durations, the MICT included prolonged exercise and monotonous nature, which may have affected the results. Furthermore, different types of training (imposed or self-paced exercise), participants' training duration, and characteristics (sex, age and training experiment) could be other influencing factors, as previously stated [15,39].

Regarding the effect of Sp-HIIT and Sp-MICT on body composition, the present study demonstrated significant favourable effects on body weight and fat percentage. No significant differences were observed between training interventions, which confirms some other findings in untrained hypertensive women [40], but are in contrast to findings in sedentary males [15]. However, none of these studies were controlled for altered nutritional habits, which is a limitation. Furthermore, the current study also demonstrated upregulated performance in the 30-15 V_{IFT} as well as VO_{2max} in Sp-HIIT compared to Sp-MICT after 8-week interventions. These findings support previous research comparing HIIT and MICT interventions with different training durations ranging from 4 to 12 weeks and in a variety of populations, such as healthy [41], recreationally active [13], and untrained young adults (15). In a similar study, Hottenrott et al. [42] reported greater increases in V_{IFT} and VO_{2max} following a 12-week HIIT intervention compared to MICT in recreationally active adults. Similarly, an 8-week HIIT intervention resulted in 11% increase in VO_{2max} in healthy men [43] a 11% improvement in young women [44], which is of similar magnitude as reported by Nybo et al [15] in healthy males and by Kristiansen et al [45] in a frail patient with coronary heart disease after 12-wks of HIIT. Contrary to these study results supporting our findings, VO_{2max} has been shown to increase 20% after a 12-week Sp-MICT training programmes compared with Sp-HIIT in inactive women [11]. From a practical point of view, our findings speak in favour that Sp-HIIT is a feasible, practical and efficient training method to improve VO_{2max} in recreationally active adults. However, there is still no consensus on which type of self-paced training intervention is optimal to improve aerobic fitness and body composition because of the lack of literature.

The present study measured circulatory oxidative stress and antioxidant markers by determining resting serum MDA, CAT, SOD, GSH and GSSG values prior and after the intervention. We demonstrated higher level of serum MDA, as well as performance in the 30-15 V_{FIT} and VO_{2max} after 8-week of Sp-HIIT compared to Sp-MICT. In contrast to our study results, some studies have found an increase in serum SOD and CAT activity and a decrease in MDA levels in untrained men after 8-week aerobic training [46,47], while our findings agree with Knez et al. [48], who found that the systemic MDA concentration increases progressively during an intervention. The analysis of circulatory MDA markers of peroxidation and antioxidant enzymes in individuals with long-term exposure to metabolic oxidative stress induced by exercise training may indicate altered fatigue pattern and, consequently, the overload of adaptive stress mechanisms and depression. Although the similar training intervention protocols in these studies, some important confounding factors such as the populations' physical activity levels, age and training intensity may explain the diverging results.

Our study demonstrated that 8-week of Sp-HIIT provoked higher increases in 30-15 V_{FIT} and VO_{2max} responses compared to Sp-MICT, but with no statistical associations between aerobic fitness level such as 30-15 V_{FIT} and VO_{2max} and circulatory antioxidant markers, which is in line with findings by others [49]. In contrast to our study results, Peserico et al. [50] have recently found strong correlations between GSSG and indicators of aerobic fitness level, such as peak velocity and 5-km running time in untrained men. In addition, an earlier study found that the systemic GSH values correlated with training volume and VO_{2max} in competitive triathletes [51]. As a result of improved VO_{2max}, increasing reactive oxygen species production is associated with increased erythrocyte glutathione peroxidase activity and glutathione concentration, both of which will protect the organism against lipid peroxidation and cell membrane damage [51]. Differences between studies in antioxidant markers and aerobic fitness level might be explained by genetic background, physical activity levels and the general participant characteristics.

The study has strengths and limitations. The applied training modalities were designed according to suggested physical activity guidelines to improve health conditions for adults, which is strength. Furthermore, the study is the first to compare the effects of self-paced training on the aerobic fitness level, psychophysiological responses and antioxidant status in young well-trained adults. However, several limitations must be addressed. The main limitation of this study is that there was a lack of direct measurement of aerobic capacity. Another important limitation is lack of control of the nutritional habits of participants, affecting physical performance in relation to body composition and blood profiles. Finally, the sample size is too small to generalize the findings and no inactive control group was included.

5. Conclusions

In conclusion, both self-paced high-intensity interval and continuous moderate-intensity training in an 8-week period demonstrated similar improvements in anthropometric performance and antioxidant status, while adaptations in 30-15 V_{FIT}, VO_{2max} and serum MDA were in favour of high-intensity interval training. However, the high-intensity interval training intervention induced higher negative changes in RPE and depression responses compared to moderate intensity continuous training.

Supplementary Materials: The following supporting information can be downloaded at the website of this paper posted on Preprints.org, Figure S1: title; Table S1: title; Video S1: title.

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