



## The impact of a single bout of high intensity circuit training on myokines' concentrations and cognitive functions in women of different age



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### ABSTRACT

The study aimed to assess effect of a single bout of high-intensity circuit training (HICT) on myokines concentration: interleukin-6 and irisin, inteleukin-10, brain-derived neurotrophic factor (BDNF), heat shock proteins (HSP27, HSP70) and cognitive functions among women participated in HICT. It also attempted evaluating whether vitamin D could have modified the effect of HICT.

Fourteen healthy, non-active women participating in the experiment were assigned to a young or middle-aged group. They performed a single session HICT using body weight as a resistance, based on the ACSM recommendations. Blood samples were taken before, one and 24 h after training. Cognitive functions were assessed before and 1 h after the HICT session. Simple statistics and effects of changes for dependent variables were determined using mixed linear modeling, and evaluated by means of magnitude-based inference (MBI).

Following a single session of HICT the young group exhibited improved concentration and spatial memory, whereas in middle-aged women these functions were attenuated. A varied tendency was also observed in the levels of myokine IL-6 and cytokine IL-10. Vitamin D was covariate for changes in cognitive functions and myokines' levels after exercise. Its concentration modified the anti-inflammatory effect of HICT, expressed in decreasing HSP70.

### 1. Introduction

The positive effect of exercise on brain structure and function is becoming more widely recognized [1]. Physical inactivity is, thus, being linked not only to an enhanced risk of morbidities like cancer or diabetes, but also deterioration of cognitive abilities [2]. Despite these findings and healthy recommendations advised, many people still find it challenging to exercise regularly due to “the lack of time”. Consequently, in recent years, researchers have turned to an interval training as a time-saving, effective form of physical activity, which induces physiological adaptations characteristic of a moderate-intensity continuous training [3,4] not only in healthy subjects but also among aging patients with diverse dysfunctions [5]. The most investigated and popular interval protocols include the high-intensity interval training (HIIT; ‘near maximal’ efforts) and sprint interval training (SIT; ‘supramaximal’ efforts). However, interactions between intensity and

duration of interval protocols have not been thoroughly explored yet. Some investigations have focused on the high-intensity circuit training (HICT) as a way of increasing the intensity of exercise and inducing healthy effects at the same time [6]. HICT can use an external resistance workload, high-intensity interval resistance training (HIRT) [7], or apply own's body weight [8]. The last option can be performed anywhere, not necessarily at the gym; therefore, it meets a growing interest in home-based exercise programmes [9].

Schulz and co-workers emphasized that high interval training HIT represents an effective and preferred intervention for elevating brain-derived neurotrophic factor (BDNF) levels and potentially, promoting brain health and plasticity; however, the authors did not specify what the most effective HIT program inducing that same effect would look like [10]. BDNF is essential for synaptic plasticity, hippocampal function and learning [11]. Several studies suggested that physical or mental activity may stimulate cognitive benefits reflected in an

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enhanced BDNF signaling [12,13]. BDNF is found to be an important factor not only in neurobiology, but also in central as well as peripheral metabolism [14] due to its ability to easily cross the blood-brain barrier [15]. It is also known the skeletal muscle would produce BDNF in response to exercise [16], but it is currently believed that its impact is locally restricted to motor units and muscle cells directly [17]. Still, other myokines released in response to exercise are considered to act as signals stimulating BDNF synthesis [18]. Research implied that the link between exercise and BDNF expression is moderated by irisin [19], the part of FNDC5, membrane protein as a newly identified hormone that is cleaved and secreted in response to physical activity. A 30-day voluntary, free running-wheel exercise led to an increase in FNDC5 in mice hippocampus, which in turn enhanced expression of BDNF [20]. The link between irisin and an increase in BDNF concentration confirms the neuroprotective effects of exercise [1]. Still, data directly evaluating the impact of exercise, its time duration and intensity on irisin, BDNF and cognitive function are unclear.

The available data point out to the pro-inflammatory cytokine TNF $\alpha$  as one of the factors, which can modulate BDNF synthesis [21,22]. Consequently, the inflammation induces Heat Shock Protein (HSP) synthesis, which regulates specific stress-responsive signaling pathways [23]. Among the subset of stress-responsive proteins, assessing HSP27 and HSP70 is considered to be a new approach to monitoring exercise and adaptive mechanisms [24]. Moreover, data published by Heck and co-workers revealed that HSP70 acts as a novel fatigue signaling factor, sent from the immune system to the brain [25]. They noted that the release of eHSP70 from lymphocytes during high-load exercise bouts may contribute to fatigue sensation. The link between HSP, BDNF concentration and cognitive functions is not, however, fully understood. At the same time, the inflammatory effect of exercise may be regulated by the concentration of vitamin D (25(OH)D3) due to its well-documented anti-inflammatory effect [26]. Due to the many essential roles of vitamin D within the body (protein synthesis, muscle function, cellular growth and regulation of skeletal muscle), it was suggested that physical performance may be influenced by the serum vitamin D status [27]. Hence we assumed, that vitamin D concentration would modify the muscle secretory function.

Body weight training appeared for the first time in the American College of Sports Medicine (ACSM) trends` survey in 2013 at no. 3 and has since advanced to no. 2 in 2017 forecast [28]. There are no published studies examining the effect of interval body-weight training on myokines` concentrations: irisin, IL-6 or BDNF. The way it affects cognitive functions in the human brain, thus, remains unclear. In light of discussed developments, the aim of our study was to assess the impact of a single bout of high intensity circuit training (HICT) on irisin, inflammation markers, HSP, BDNF concentrations and cognitive functions among young and middle-aged women. In addition, we assessed the role of vitamin D (25(OH)D3) in moderating inflammatory response to exercise and myokines' cross-talk.

## 2. Materials and methods

### 2.1. Subjects

Fourteen healthy women, who had not exercised regularly for at least 1 year, only occasionally or spontaneously, participated in the experiment. The participants were assigned either to the young group YG ( $n = 8$ ,  $22.7 \pm 3$  years) or middle-aged group MG ( $n = 6$ ,  $41.7 \pm 4$  years). All participants were non-smokers, free from any diseases and not taking any medications. Women were tested in the mid-follicular phase of their menstrual cycles. Written, informed consent was obtained from all subjects. All study procedures were approved by the Bioethical Committee of the Regional Medical Society in Gdansk KB-14/17. One week prior to the start of the experiment, body composition, aerobic capacity and daily activities' assessments were performed.

### 2.2. Body composition assessment

Body mass (BM) and body composition were estimated using a multi-frequency impedance plethysmograph body composition analyser (In Body 720, Biospace, Korea). It accurately measures the amount of body water as well as body composition, including fat mass, free fat mass, skeletal muscle mass and soft lean mass. Additionally, the visceral fat area was determined and expressed in  $\text{cm}^2$ . Precision of the repeated measurements was expressed as the coefficient of variation, which was 0.6% for fat mass percentage on average [29].

### 2.3. Cardiorespiratory fitness measurement

In order to determine  $\text{VO}_{2\text{max}}$ , the participants performed a graded cycle ergometry test on a cycle ergometer with electromagnetically applied brakes (ER 900 Jaeger, Germany/Viasys Health Care). The participants were allowed a 5-min warm up period at an intensity of  $1.5 \text{ W kg}^{-1}$ , with a pedalling cadence of 60 rpm. Immediately following the warm-up, the participants began  $\text{VO}_{2\text{max}}$  testing by cycling at a workload increasing at a rate of  $25 \text{ W min}^{-1}$  until they reached the point of volitional exhaustion [30]. Breath-by-breath pulmonary gas exchange was measured (MetaMax 3B, Cortex, Germany) throughout the  $\text{VO}_{2\text{max}}$  test. The equipment was calibrated according to the manufacturer guidelines in three stages: 1) Ambient conditions calibration, including the temperature, atmospheric pressure and humidity; 2) Volume calibration, using a standard 2000 ml syringe, 3) Gas calibration of the  $\text{O}_2$  and  $\text{CO}_2$  analysers before each test using standard gases, following provided instructions.

### 2.4. Daily activity

ActiGraph GT3X accelerometer was used to establish daily physical activity of the subjects, expressed in the total amount of steps [31]. The subjects each spent one whole day of typical activities wearing the accelerometer and recording their movements.

### 2.5. Training intervention

All participants completed a single HICT session, using body weight as resistance according to the ACSM recommendations [8]. One week before the main experiment, they were introduced to the protocol, with the research assistant explaining and showing how to properly perform each exercise. From the original circuit training protocol ten exercises were selected: jumping jacks, push-up, abdominal crunch, squat, plank, triceps dip on chair, high knees/running in place, lunge, push-up with rotations, side plank. Two exercises were excluded from the original protocol due to participants' safety: a wall sit and a step-up onto a chair. Individual training sessions were performed at 09:00, one hour after the participants ate a light breakfast. Exercises were each performed for 30 s, with 10 s of transition time in-between. The entire circuit took approximately 7 min to complete and was repeated 3 times with 2-minute breaks in-between circuits. The same research assistant controlled the time for interval and recovery periods. The subjects were instructed to perform all exercises with a maximal intensity (to make as many repetitions as possible within a 30-s slot). Every participant wore a Suunto heart rate belt to control the intensity of training session.

### 2.6. Assessment of cognitive functions

Cognitive functions were assessed at baseline and 1 h after the training session. Each participant completed three computer-based tests measuring three domains: 1) the storage capacity of spatial short-term memory and of learning in spatial middle-aged memory (Corsi Block-Tapping-Test – CORSI S3), 2) cognitive control including selective attention and response inhibition based on the colour-word interference tendency (Stroop Interference Test – Stroop S8) and 3) the ability to

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