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Endurance training increases exercise-induced prostacyclin release in young, healthy men – relationship with VO_{2max}

Jerzy A. Zoladz[1], Joanna Majerczak[1], Krzysztof Duda[1], Stefan Chłopicki[2]

[1]Department of Muscle Physiology, Chair of Physiology and Biochemistry, University School of Physical Education, Al. Jana Pawła II 78, PL 31-571 Kraków, Poland

[2]Department of Experimental Pharmacology, Chair of Pharmacology, Jagiellonian University Medical College, Grzegórzecka 16, PL 31-531 Kraków, Poland

Correspondence: Jerzy A. Zoladz, e-mail: Jerzy.Zoladz@awf.krakow.pl

Abstract:
In the present study, we evaluated the effect of 5 weeks of moderate-intensity endurance training on the basal and exercise-induced systemic release of prostacyclin (PGI_2), as assessed by plasma 6-keto-$PGF_{1\alpha}$ concentration. Twelve physically active young men with the following characteristics participated in this study (the mean ± SD): age, 22.7 ± 2.0 years; body mass, 76.8 ± 8.9 kg; BMI, 23.48 ± 2.17 kg × m^{-2}; and maximal oxygen uptake (VO_{2max}), 46.1 ± 4.0 ml × kg^{-1} × min^{-1}. Plasma 6-keto-$PGF_{1\alpha}$ concentrations were measured in venous blood samples taken prior to the exercise and at exhaustion (at VO_{2max}) before and after completing the training protocol. On average, the training resulted in a significant increase in VO_{2max} ($p = 0.03$), power output at VO_{2max} ($p = 0.001$) and a significant increase ($p = 0.05$) in the net-exercise-induced increase in plasma 6-keto-$PGF_{1\alpha}$ concentration (Δ6-keto-$PGF_{1\alpha}$ i.e., the difference between the end-exercise and pre-exercise 6-keto-$PGF_{1\alpha}$ concentrations). No effect of training on the basal PGI_2 concentration was found. Interestingly, within the study sample ($n = 12$), two subgroups could be defined with a differential pattern of response with respect to Δ6-keto-$PGF_{1\alpha}$ concentrations. In one subgroup ($n = 7$), a significant increase in Δ6-keto-$PGF_{1\alpha}$ concentration after training was found ($p < 0.02$) (responders). This enhancement in the exercise-induced PGI_2 release was accompanied by a significant ($p < 0.05$) increase in VO_{2max} after training. In contrast, in another subgroup ($n = 5$), there was no observed effect of training on the Δ6-keto-$PGF_{1\alpha}$ concentration and the VO_{2max} after training (non-responders). In both of these subgroups, training did not influence the basal PGI_2 concentration.
In conclusion, the endurance training resulted in the adaptive augmentation of the systemic release of PGI_2 in response to exercise, which plays a role in the training-induced increase in VO_{2max} in young, healthy men. The impairment of the training-induced augmentation of PGI_2 release in response to exercise demonstrated in the non-responders subgroup may predispose them to increased cardiovascular risk during vigorous exercise.

Key words:
endurance training, exercise, maximal oxygen uptake, power output, prostacyclin

Abbreviations: BMI – body mass index, CO – carbon monoxide, EDHF – endothelium-derived hyperpolarizing factor, LT – lactate threshold, NO – nitric oxide, NR – non-responders group, PGI_2 – prostacyclin, PO at VO_{2max} – power generated at the VO_{2max}, POST – after training, PRE – before training, R – responders group, VO_{2max} – maximal oxygen uptake

Introduction

Proper endothelial function is essential to maintaining vascular wall health, and the endothelium-dependent vasoprotective mechanisms activated by exercise

seem to play an important role in the exercise-related prevention of cardiovascular diseases. The endothelium produces a number of vasoprotective mediators, including nitric oxide (NO), prostacyclin (PGI_2), endothelium-derived hyperpolarizing factor (EDHF), carbon monoxide (CO) and other mediators, which exert anti-thrombotic, anti-inflammatory and vasoprotective actions [1, 11, 18]. Currently, the effects of exercise on NO-dependent responses are the most widely studied. Numerous studies have demonstrated that short-term exercise and training induce improvement in endothelial, NO-dependent function [16]; this response may provide vasodilator, antioxidative, antiadhesive, antiproliferative and antiatherosclerotic effects [26]. PGI_2 is a potent, endogenous inhibitor of platelet activity, a stimulator of thrombomodulin a vasodilator and a potent antiatherosclerotic molecule [17, 19] that could also play a major role in the vascular adaptation to exercise. However, the endothelial PGI_2-dependent response to exercise is not well understood. It was previously reported that a single bout of exercise increased the concentration of the PGI_2 metabolite, 6-keto-$PGF_{1\alpha}$, in urine [8, 25, 47], in blood [3, 28, 38, 53] or in the interstitial fluid of muscles [14, 22]. Moreover, it was reported that an 8-week endurance training program resulted in a significant increase in the urinary metabolite prostacyclin (2,3-dinor-6-keto-prostaglandin $F_{1\alpha}$) in untrained boys [43]. However, to our best knowledge, no data exist regarding the effect of training on the exercise-induced release of PGI_2 in humans. Therefore, it is unknown whether this response is modulated by training.

Interestingly, we demonstrated that the systemic release of PGI_2 during maximal exercise positively correlates with the maximal oxygen uptake (VO_{2max}) in young, healthy men [53], suggesting that the vascular capacity to release PGI_2 during maximal exercise might play a significant role in the mechanism determining VO_{2max} in humans.

It is well known that a relatively short period of endurance training (weeks – months) increases the VO_{2max} and the exercise tolerance of humans [15, 34, 50]. However, it was also reported that the magnitude of the training-induced increase in VO_{2max} varies substantially among subjects and seems to be partially genetically determined [9, 13, 34]. For example, it was demonstrated by Prud'homme et al. [34] that 20 weeks of endurance training resulted, on average, in a 20% increase in VO_{2max}; however, in some subjects, a negligible or no increase in VO_{2max} was observed after training [9].

If the systemic release of PGI_2 does indeed play a significant role in the mechanism determining VO_{2max} in humans, then the training-induced increase in VO_{2max} should be accompanied by an increase in the magnitude of the exercise-induced PGI_2. Accordingly, in the present study, we hypothesized that the training-induced increase in the VO_{2max} would be accompanied by an increase in the magnitude of systemic PGI_2 release during maximal exercise in young, healthy men.

Materials and Methods

Subjects characteristics

Twelve non-smoking men (the mean ± SD: age, 22.7 ± 2.0 years; body mass, 76.8 ± 8.9 kg; height, 180.7 ± 5.6 cm; BMI, 23.48 ± 2.17 kg × m^{-2}; VO_{2max}, 3,521 ± 334 ml × min^{-1}, i.e., 46.1 ± 4.0 ml × kg^{-1} × min^{-1}) participated in this study. All procedures were approved by the Local Ethics Committee and performed according to the Declaration of Helsinki. Subjects provided informed, written consent and were aware of the aims of the study.

Exercise protocol

The incremental exercise test was performed on the cycloergometer Ergo-Line GmbH & Co. KG 800s (Bitz, Germany). Before the test, a 6-min resting period was allowed to determine the resting stage of the cardiorespiratory parameters and to withdraw the blood samples. The exercise test started at a power output (PO) of 30 W, followed by gradual increases amounting to 30 W every 3 min and continued until exhaustion [52]. The pedaling rate during the test amounted to 60 rev × min^{-1}. The PO at which the subjects reached VO_{2max} was termed the PO at VO_{2max}. The incremental exercise test was performed twice: once at 3 days before training and once at 3 days after the completion of endurance training.

Gas exchange variables

Gas exchange variables were measured continuously, breath-by-breath, using the Oxycon Champion (Mijnhardt BV, Bunnik, The Netherlands). Measurements began 6 min prior to exercise and lasted until the test was stopped. Before and after each test, gas analyzers were calibrated with certificated calibration gases as previously described by Zoladz et al. [53].

Blood sampling

Blood samples were taken using an Abbot Int-Catheter, Ireland (18G/1.2 × 45 mm), inserted into the antecubital vein approximately 15 min prior to the onset of the exercise. The catheter was connected to an extension set using a "T" Adapter SL from Abbot, Ireland (the tube was 10 cm in length). Immediately before taking each blood sample, 1 ml of blood was taken to eliminate blood from the catheter and the T-set. Blood samples for plasma lactate concentrations were taken prior to the exercise test, at the end of each step of the incremental exercise (the last 15 s before increased PO) and at the end of the exercise protocol. Blood samples for the measurement of PGI_2 metabolite (6-keto-$PGF_{1\alpha}$) were taken prior to the exercise at rest and at the end of the exercise protocol (at exhaustion). The magnitude of exercise-induced increase in plasma 6-keto-$PGF_{1\alpha}$, defined as the difference between the end-exercise and pre-exercise plasma concentrations of 6-keto-$PGF_{1\alpha}$ (Δ6-keto-$PGF_{1\alpha}$), was considered to be a reliable index of exercise-induced PGI_2 release. In theory, Δ6-keto-$PGF_{1\alpha}$ could be determined not only by PGI_2 production but also by the rate of PGI_2 degradation and the rate of 6-keto-$PGF_{1\alpha}$ elimination. However, it seems unlikely that an alteration in the rate of degradation of PGI_2 or in the elimination of 6-keto-$PGF_{1\alpha}$ was responsible for the increase in 6-keto-$PGF_{1\alpha}$ during a single exercise of maximal intensity, as applied in our experimental setting. Therefore, the exercise-induced increase in 6-keto-$PGF_{1\alpha}$ concentration was attributed to the exercise-induced PGI_2 release.

Plasma 6-keto-$PGF_{1\alpha}$ measurements

For the determination of 6-keto-$PGF_{1\alpha}$ concentration, blood samples were collected in Eppendorf tubes with 10 μM indomethacin and 1 mM EDTA (final concentration) and immediately spun for 5 min at 2,000 × g to obtain plasma. Plasma samples were stored at –70°C. The concentration of 6-keto-$PGF_{1\alpha}$ in plasma prior to the exercise test and at the end of the exercise protocol were assayed using commercially available enzyme immunoassay kits (R&D Systems, Inc., MN, USA) and expressed in pg × ml^{-1}.

Plasma lactate measurements

The samples for plasma lactate concentration determinations (0.5 ml each) were placed in 1.8 ml Eppendorf tubes containing 1 mg ammonium oxalate and 5 mg sodium fluoride, mixed for approximately 20 s and then centrifuged. The obtained plasma samples (200 μl) were stored at –32°C for further analysis of lactate concentration ($[La^-]_{pl}$) using an automatic analyzer Vitros 250 Dry Chemistry System, Kodak (Rochester, NY, USA).

Lactate treshold

In the present study, the lactate threshold (LT) was determined for the purpose of objective scaling of the training intensity (see above). The LT was defined as the highest PO above which plasma lactate concentration [La^-] showed a sustained increase of more than 0.5 mmol × L^{-1} × $step^{-1}$ [54].

Endurance training program

A five-week endurance training program was performed on cycloergometers (Monark 874 E) at pedaling rates of 60 rev × min^{-1} according to two different protocols. The continuous endurance cycling protocol was applied for 40 min on Tuesdays and Fridays at the PO corresponding to 90% of oxygen consumption, which was measured at the previously determined lactate threshold (90% VO_2 at LT). The intermittent endurance cycling protocol was performed on Mondays and Thursdays and was comprised of 6 min of cycling without resistance (unloaded cycling) and 3 min of cycling at the PO corresponding to 50% Δ, which was calculated for each subject according to the following formula: 50% Δ = PO at LT + 0.5 (PO_{max}–PO_{LT}) [4]. This intermittent bouts of cycling exercise were repeated four times during each session of this kind and ended with 4 min of unloaded cycling. There was no training on Wednesdays, Saturdays and Sundays. In total, each volunteer performed, on average, 20.8 ± 0.56 training sessions for 13.9 ±

0.37 h. The training workload was predominantly of moderate intensity because 85% of the workload (expressed in time) was performed below the LT and only 15% was performed above the LT (at 50% Δ, see above). The scaling of the training intensity was based on the principle that the primary training workload should be performed in the moderate exercise-intensity domain [49] in order to recruit predominantly type I muscle fibers [42]. However, some of the training workloads were planned to be performed in the heavy-intensity domain [49] in order to additionally recruit type II muscle fibers [42].

Statistics

The results are expressed as the means ± SD. Significance was set at $p < 0.05$. Statistical significance for paired samples was tested using the nonparametric Wilcoxon signed-rank test; non-asymptotic, exact, two-sided p-values are presented. The statistics were done using the statistical packet StatXact 6.1 and STATISTICA 7.1.

Results

The effects of training on exercise-induced PGI_2 release: the responders (R) and non-responders (NR)

The results of this study are presented for the total subjects (n = 12) and also for the R (n = 7) group and the NR (n = 5) group. The groups R and NR were established based on the effect of the performed training on the end-exercise plasma 6-keto-$PGF_{1\alpha}$ concentrations. Group R includes the subjects with an increased end-exercise plasma 6-keto-$PGF_{1\alpha}$ concentration after training, and the group NR includes the subjects with unchanged or decreased end-exercise plasma 6-keto-$PGF_{1\alpha}$ concentration after training.

The maximal oxygen uptake (VO_{2max})

The VO_{2max} prior to training for the total subjects was 3,521 ± 334 ml × min^{-1}; after training, it increased significantly to 3,624 ± 363 ml × min^{-1} ($p = 0.03$). In R group, the pre-training VO_{2max} was 3,477 ± 231 ml × min^{-1}; after training, it increased significantly ($p = 0.047$) to 3,614 ± 199 ml × min^{-1}. In group NR, pre-training VO_{2max} was 3,581 ± 468 ml × min^{-1}; after training, it increased to 3,637 ± 549 ml × min^{-1} ($p = 0.44$).

The maximal power output reached at the maximal oxygen uptake (PO at VO_{2max})

The PO at VO_{2max} reached before training for the total subjects amounted to 257.7 ± 7.1 W. The training resulted in a significant ($p = 0.001$) increase to 279.6 ± 7.0 W. The PO at VO_{2max} in the R group before training was 254.9 ± 14.1 W; after training, it increased to 279.7 ± 9.9 W ($p = 0.03$). In the NR group, the PO at VO_{2max} before training was 261.6 ± 36.7 W; after training, a tendency ($p = 0.06$) towards a significant increase in the PO at VO_{2max} (279.4 ± 38.3 W) was found.

Basal plasma 6-keto-$PGF_{1\alpha}$ concentration before and after training

Before training, the basal plasma 6-keto-$PGF_{1\alpha}$ concentration for the total subjects was 46.8 ± 13.5 pg × ml^{-1}. After 5 weeks of endurance training, it was unaffected ($p = 0.38$), displaying a basal concentration of 6-keto-$PGF_{1\alpha}$ at 41.2 ± 11.1 pg × ml^{-1}. After training, the average end-exercise plasma 6-keto-$PGF_{1\alpha}$ concentration in this group amounted to 95.8 ± 22.9 pg × ml^{-1} as compared with the pre-training state of 62.7 ± 11.9 pg × ml^{-1}, but this difference did not reach statistical significance ($p = 0.20$). However, the exercise-induced increase in the 6-keto-$PGF_{1\alpha}$ (expressed as Δ6-keto-$PGF_{1\alpha}$, i.e., max-basal) after training was significantly higher than before training ($p = 0.05$) (see Fig. 1).

Varied effects of the endurance training on the plasma 6-keto-$PGF_{1\alpha}$ concentration: responders and non-responders

There was a pronounced variability in the magnitude of response in the plasma 6-keto-$PGF_{1\alpha}$ levels at the end of the incremental exercise. In 7 subjects (group R), a significant ($p = 0.016$) increase in the end-exercise plasma 6-keto-$PGF_{1\alpha}$ was noted when compared to basal level (see Fig. 2A). In 5 subjects (group NR), however, there was no change ($p = 0.63$) in the exercise-induced increase in the 6-keto-$PGF_{1\alpha}$ after

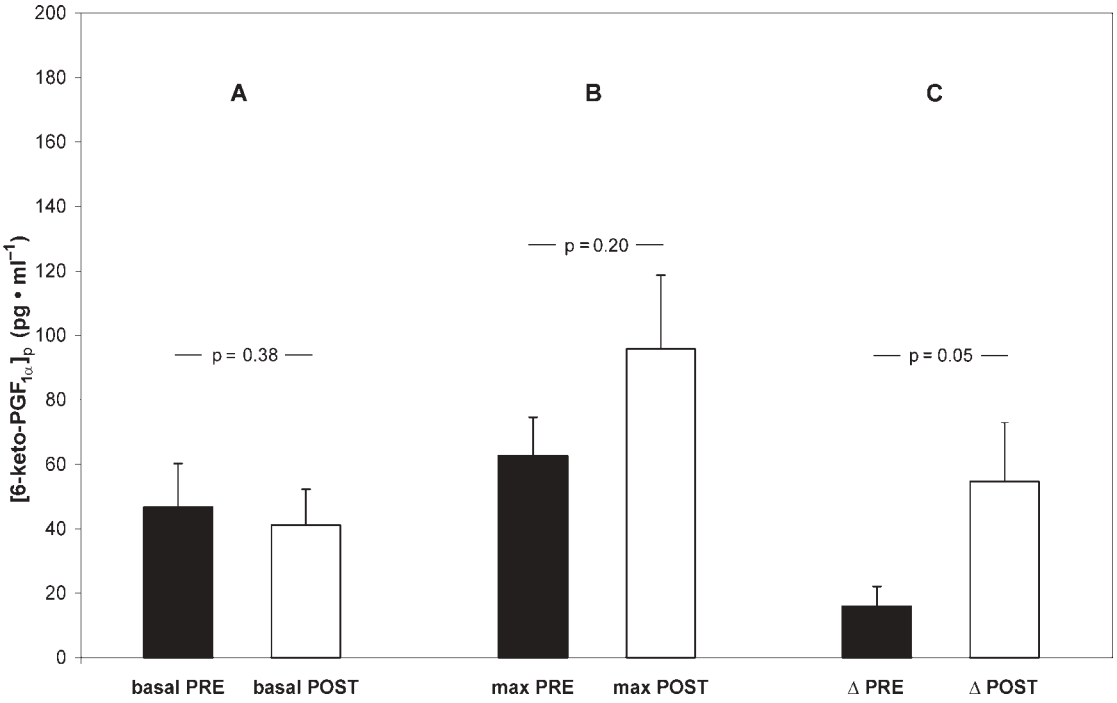

Fig. 1. The plasma 6-keto-$PGF_{1\alpha}$ concentrations for the total subjects (n = 12). Panel **A**: basal before (basal PRE) and after training (basal POST). Panel **B**: at the VO_{2max} reached during the incremental test, performed before training (max PRE), and at the VO_{2max} reached after training (max POST). Panel **C**: the difference in the plasma 6-keto-$PGF_{1\alpha}$ concentrations between its VO_{2max} level minus its basal concentration before (Δ PRE) and after training (Δ POST)

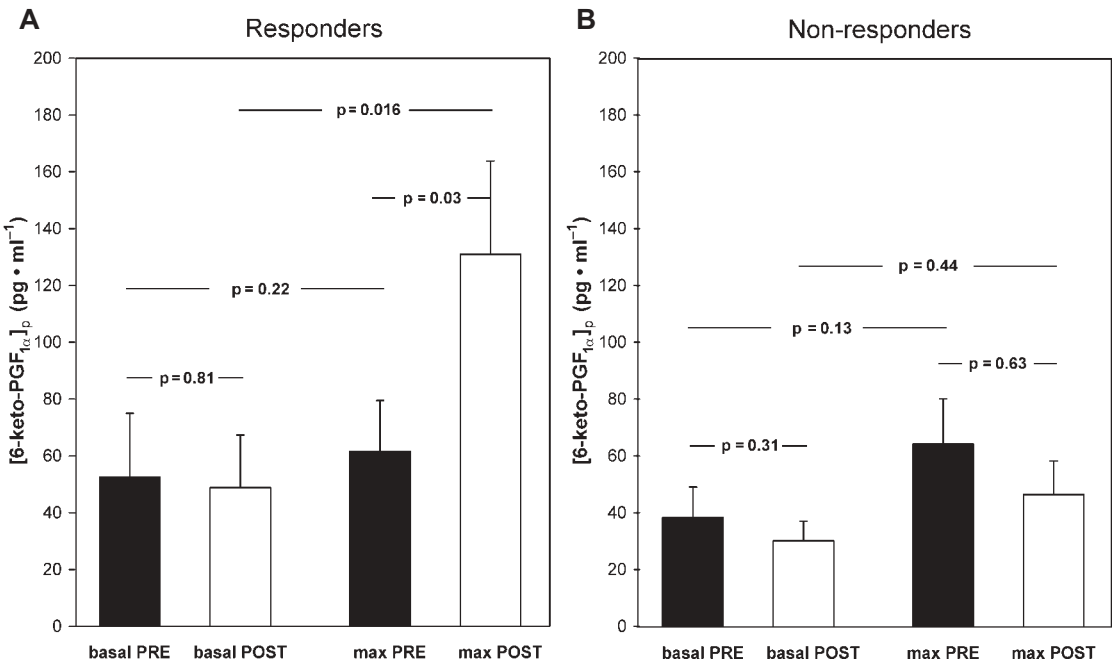

Fig. 2. Plasma 6-keto-$PGF_{1\alpha}$ concentration in the group of responders (n = 7) (left) and in the group of non-responders (n = 5) (right) at rest (basal) before (PRE) and after training (POST) as well as at the VO_{2max} (max)

training (see Fig. 2B). Interestingly, the basal 6-keto-$PGF_{1\alpha}$ in group R was not significantly affected by training ($p = 0.81$). Before training, the basal 6-keto-$PGF_{1\alpha}$ amounted to 52.5 ± 22 pg × ml^{-1}, and it was 48.8 ± 18 pg × ml^{-1} after training. In group NR, basal 6-keto-$PGF_{1\alpha}$ was also not significantly ($p = 0.31$) affected by training. Before training, basal 6-keto-$PGF_{1\alpha}$ amounted to 38.5 ± 10 pg × ml^{-1} *vs*. 30.5 ± 6 pg × ml^{-1} after training.

Discussion

The primary and original finding of the present study is that moderate-intensity endurance training lasting five weeks resulted in a significant increase ($p = 0.05$) in the systemic release of PGI_2 during maximal incremental cycling exercise in young, healthy men (see Fig. 1). This effect was accompanied by a significant ($p = 0.03$) increase in the VO_{2max} and by a significant ($p = 0.001$) increase in the PO at VO_{2max}. The systemic release of PGI_2 in this study was expressed as the Δ6-keto-$PGF_{1\alpha}$ i.e., the plasma 6-keto-$PGF_{1\alpha}$ concentration at exhaustion (at VO_{2max}) minus the plasma 6-keto-$PGF_{1\alpha}$ concentration at rest [53].

Surprisingly, even among the small group of subjects enrolled in this study, we identified responders (R) and non-responders (NR) subgroups who displayed improvement or no improvement in exercise-induced PGI_2 release following endurance training, respectively. In the R group ($n = 7$), we observed a significant increase ($p = 0.016$) in the exercise-induced release of PGI_2 (Δ6-keto-$PGF_{1\alpha}$) after training (see Fig. 2). This enhancement in the exercise-induced PGI_2 release was accompanied by a significant ($p < 0.05$) increase in VO_{2max} after training. In contrast, in the NR ($n = 5$) group, there was no observed effect of training on the Δ6-keto-$PGF_{1\alpha}$ ($p = 0.63$) and the VO_{2max} after training ($p = 0.44$). In both of these subgroups, training did not influence the basal PGI_2 concentration.

Various sensitivities to training that affect VO_{2max} are well documented but not completely understood [9, 13]. Surprisingly, in the present study, we observed for the first time (to our knowledge) that the magnitude of response to endurance training, expressed by an increase in VO_{2max}, is related to the magnitude of systemic PGI_2 release during maximal exercise. The relationship between the training-induced increase in the systemic release of PGI_2 during exercise and the increase in the VO_{2max} can be explained by the fact that VO_{2max} in healthy men is primarily determined by the amount of oxygen delivered to the working muscle (for a review see [2, 6, 37, 41]). PGI_2 can increase oxygen delivery to the working muscle in various ways [52], including the enhancement of coronary vasodilatation and the increase of cardiac output [29, 31, 35] during maximal exercise. This increase in cardiac output results from improved function in the right heart chamber by lowering the pulmonary arterial pressure, which increases during exercise [21, 23], and from improved gaseous exchange in the lungs during exercise by limiting alveolar edema formation [40].

Interestingly, the results of the present study showing an increase in VO_{2max} after training only in the responders subgroup (see Fig. 2 and 3) clearly support the hypothesis that the exercise-induced increase in PGI_2 is an important factor determining VO_{2max}. This notion is in accordance with our recent findings [53] showing a significant positive correlation between Δ6-keto-$PGF_{1\alpha}$ and the VO_{2max} (see Fig. 2 therein).

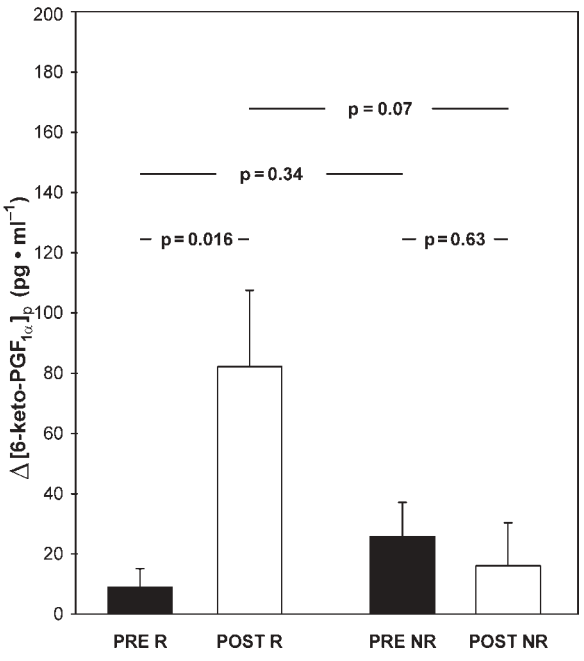

Fig. 3. The differences in the plasma 6-keto-$PGF_{1\alpha}$ concentrations determined at VO_{2max} and at basal condition. Δ6-keto-$PGF_{1\alpha}$ before (PRE) and after training (POST) in the responders (R) ($n = 7$) and in the non-responders (NR) ($n = 5$) groups

It is worth noting that the anti-platelet activity of PGI_2 may play an important role during exercise. Vigorous exercise causes platelet activation and increases platelet-platelet and platelet-leukocyte aggregates [24, 27], which may limit microcirculation perfusion during exercise and increase the cardiovascular risk of exercise [5]. Several studies have demonstrated that PGI_2 or PGI_2 analogues increase exercise capacity [7, 10, 46, 48]. This increase might be due to the anti-platelet effects of PGI_2. Iloprost (PGI_2 analog) consistently prolonged exercise duration and reduced platelet aggregation at peak exercise, suggesting that the anti-platelet effects of PGI_2 may account for myocardial or skeletal blood perfusion during vigorous exercise and thus determine the exercise capacity in these patients.

In contrast, it is well documented that individuals with poor physical capacity, such as elderly people, patients who have suffered myocardial infarction and diabetics are characterized by a poor ability to release PGI_2 during exercise [25, 36, 45, 51]. Moreover, this group of people is at high cardiovascular risk during exercise [5]. Accordingly, the exercise-induced systemic release of PGI_2 may determine exercise performance while also limiting the cardiovascular risk of exercise.

Taking the above facts into account, the failure of training to improve the endothelial PGI_2 response to exercise in the NR subgroup may not only translate into worse exercise performance and lower VO_{2max} (likely related to the limitation in perfusion of the microcirculation during exercise [12, 20, 30, 32, 39] or by other mechanisms (see above)) but may also lead to the excessive platelet activation that determines the cardiovascular risk of exercise. Indeed, PGI_2 has anti-platelet, vasculoprotective, cardioprotective and anti-atherogenic properties (see e.g. [17, 19]), and the impaired exercise-induced PGI_2 release may be insufficient to hamper exercise-induced pro-thrombotic platelet activation [33]. Furthermore, the magnitude of the training-induced activation of PGI_2-dependent vasoprotective mechanism may provide a surrogate marker for exercise-related cardiovascular risk. Obviously, this hypothesis needs to be tested in a larger study. Interestingly, it was recently reported that an absence of exercise capacity improvement after an exercise training program among patients with chronic heart failure provided a strong prognostic value for adverse cardiac events, independent of classical predictive factors such as left ventricular ejection fraction [44].

In conclusion, we suggest that a training-induced adaptive increase in PGI_2 dependent-response to acute exercise plays a decisive role in the training-induced increase in VO_{2max} in young, healthy men. Further, this increase may also limit the cardiovascular risks of vigorous exercise. We also postulate that the assessment of vascular capacity to release PGI_2 during a single bout of maximal exercise in athletes and in people undertaking vigorous physical activity programs may prove useful in evaluating the cardiovascular hazard of vigorous exercise.

Conflict of interest:
There is no conflict of interest.

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References:

1. Aird WC: Endothelium in health and disease. Pharmacol Rep, 2008, 60, 139–143.
2. Andersen P, Saltin B: Maximal perfusion of skeletal muscle in man. J Physiol, 1985, 366, 233–249.
3. Barrow SE, Dollery CT, Heavey DJ, Hickling NE, Ritter JM, Vial J: Effect of vasoactive peptides on prostacyclin synthesis in man. Br J Pharmacol, 1986, 87, 243–247.
4. Barstow TJ, Jones AM, Nguyen PH, Casaburi R: Influence of muscle fiber type and pedal frequency on oxygen uptake kinetics of heavy exercise. J Appl Physiol, 1996, 81, 1642–1650.
5. Bartsch P: Platelet activation with exercise and risk of cardiac events. Lancet, 1999, 354, 1747–1748.
6. Bassett DT, Holewy ET: Limiting factors for maximum oxygen uptake and determinants of endurance performance. Med Sci Sports Exerc, 2000, 32, 70–84.
7. Blumberg FC, Riegger GA, Pfeifer M: Hemodynamic effects of aerosolized iloprost in pulmonary hypertension at rest and during exercise. Chest, 2002, 121, 1566–1571.
8. Boger RH, Bode-Boger SM, Schroder EP, Tsikas D, Frolich JC: Increased prostacyclin production during exercise in untrained and trained men: effect of low-dose aspirin. J Appl Physiol, 1995, 78, 1832–1838.
9. Bouchard C, Rankinen T: Individual differences in response to regular physical activity. Med Sci Sports Exerc, 2001, 33, Suppl 6, S446–S451.
10. Bugiardini R, Galvani M, Ferrini D, Gridelli C, Mari L, Puddu P, Lenzi S: Effects of iloprost, a stable prostacyclin analog, on exercise capacity and platelet aggregation in stable angina pectoris. Am J Cardiol, 1986, 58, 453–459.

11. Chłopicki S, Gryglewski RJ: Angiotensin converting enzyme (ACE) and HydroxyMethylGlutaryl-CoA (HMG-CoA) reductase inhibitors in the forefront of pharmacology of endothelium. Pharmacol Rep, 2005, 57, Suppl, 86–96.
12. Ciuffetti G, Sokola E, Lombardini R, Pasqualini L, Pirro M, Mannarino E: The influence of iloprost on blood rheology and tissue perfusion in patients with intermittent claudication. Kardiol Pol, 2003, 59, 197–204.
13. Dionne FT, Turcotte L, Thibault MC, Boulay MR, Skinner JS, Bouchard C: Mitochondrial DNA sequence polymorphism, VO_{2max}, and response to endurance training. Med Sci Sports Exerc, 1993, 25, 766–774.
14. Frandsen U, Bangsbo J, Langberg H, Saltin B, Hellsten Y: Inhibition of nitric oxide synthesis by systemic N_G-monomethyl-L-arginine administration in humans: effects on interstitial adenosine, prostacyclin and potassium concentrations in resting and contracting skeletal muscle. J Vasc Res, 2000, 37, 297–302.
15. Fukuoka Y, Grassi B, Conti M, Guiducci D, Sutti M, Marconi C, Cerretelli P: Early effects of exercise training on on- and off-kinetics in 50-year-old subjects. Pflugers Arch, 2002, 443, 690–697.
16. Green DJ, Maiorana A, O'driscoll G, Taylor R: Effect of exercise training on endothelium-derived nitric oxide function in humans. J Physiol, 2004, 561, 1–25.
17. Grosser T, Fries S, Fitzgerald GA: Biological basis for the cardiovascular consequences of COX-2 inhibition: therapeutic challenges and opportunities. J Clin Invest, 2006, 116, 4–15.
18. Gryglewski RJ: Prostacyclin among prostanoids. Pharmacol Rep, 2008, 60, 3–11.
19. Gryglewski RJ: Prostaglandins, platelets, and atherosclerosis. CRC Crit Rev Biochem, 1980, 7, 291–338.
20. Hill LL, Pearl RG: Combined inhaled nitric oxide and inhaled prostacyclin during experimental chronic pulmonary hypertension. J Appl Physiol, 1999, 86, 1160–1164.
21. Humbert M, Sitbon O, Simonneau G: Treatment of pulmonary arterial hypertension. (Review). N Engl J Med, 2004, 351, 1425–1436.
22. Karamouzis M, Karamouzis I, Vamvakoudis E, Ampatzidis G, Christoulas K, Angelopoulou N, Mandroukas K: The response of muscle interstitial prostaglandin E_2 (PGE_2), prostacyclin I_2 (PGI_2) and thromboxane A_2 (TXA_2) levels during incremental dynamic exercise in humans determined by in vivo microdialysis. Prostaglandins Leukot Essent Fatty Acids, 2001, 64, 259–263.
23. Kerbaul F, Brimioulle S, Rondelet B, Dewachter C, Hubloue I, Naeije R: How prostacyclin improves cardiac output in right heart failure in conjunction with pulmonary hypertension. Am J Respir Crit Care Med, 2007, 175, 846–850.
24. Kestin AS, Ellis PA, Barnard MR, Errichetti A, Rosner BA, Michelson AD: Effect of strenuous exercise on platelet activation state and reactivity. Circulation, 1993, 88, 1502–1511.
25. Koivisto VA, Jantunen M, Sane T, Helve E, Pelkonen R, Viinikka L, Ylikorkala O: Stimulation of prostacyclin synthesis by physical exercise in type I diabetes. Diabetes Care, 1989, 12, 609–614.
26. Kojda G, Hambrecht R: Molecular mechanisms of vascular adaptations to exercise. Physical activity as an effective antioxidant therapy? Cardiovasc Res, 2005, 67, 187–197.
27. Li N, He S, Blomback M, Hjemdahl P: Platelet activity, coagulation, and fibrinolysis during exercise in healthy males: effects of thrombin inhibition by argatroban and enoxaparin. Arterioscler Thromb Vasc Biol, 2007, 27, 407–413.
28. Mehta J, Mehta P, Horalek C: The significance of platelet-vessel wall prostaglandin equilibrium during exercise-induced stress. Am Heart J, 1983, 105, 895–900.
29. Merkus D, Sorop O, Houweling B, Boomsma F, Van Den Meiracker AH, Duncker DJ: NO and prostanoids blunt endothelin-mediated coronary vasoconstrictor influence in exercising swine. Am J Physiol Heart Circ Physiol, 2006, 291, H2075–H2081.
30. Muller B, Krais T, Sturzebecher S, Witt W, Schillinger E, Baldus B: Potential therapeutic mechanisms of stable prostacyclin (PGI_2)-mimetics in severe peripheral vascular disease. Biomed Biochim Acta, 1988, 47, S40–S44.
31. Nosaka S, Hashimoto M, Sasaki T, Ku K, Saitoh Y, Yamauchi M, Tanabe Y et al.: The effects of transmural pressure on prostacyclin release from porcine endocardial endothelial cells – comparison with vascular endothelial cells. Pflugers Arch, 1997, 433, 848–850.
32. Pasqualini L, Pirro M, Lombardini R, Ciuffetti G, Dragani P, Mannarino E: A human model of platelet-leucocyte adhesive interactions during controlled ischaemia in patients with peripheral vascular disease. J Clin Pathol, 2002, 55, 946–950.
33. Petidis K, Douma S, Doumas M, Basagiannis I, Vogiatzis K, Zamboulis C: The interaction of vasoactive substances during exercise modulates platelet aggregation in hypertension and coronary artery disease. BMC Cardiovasc Disord, 2008, 8, 11.
34. Prud'homme D, Bouchard C, Leblanc C, Landry F, Fontaine E: Sensitivity of maximal aerobic power to training is genotype-dependent. Med Sci Sports Exerc, 1984, 16, 489–493.
35. Raczka E, Quintana A: Effects of intravenous administration of prostacyclin on regional blood circulation in awake rats. Br J Pharmacol, 1999, 126, 1325–1332.
36. Rasmanis G, Vesterqvist O, Green K, Edhag O, Henriksson P: Prostacyclin production in myocardial infarction in the acute phase and during follow-up. J Intern Med, 1991, 229, 135–141.
37. Richardson RS, Saltin B: Human muscle blood flow and metabolism studied in the isolated quadriceps muscles. Med Sci Sports Exerc, 1998, 30, 28–33.
38. Ritter JM, Barrow SE, Blair IA, Dollery CT: Release of prostacyclin in vivo and its role in man. Lancet, 1983, 1, 317–319.
39. Rocca GD, Coccia C, Pompei L, Ruberto F, Venuta F, De Giacomo T, Pietropaoli P: Hemodynamic and oxygenation changes of combined therapy with inhaled nitric oxide and inhaled aerosolized prostacyclin. J Cardiothorac Vasc Anesth, 2001, 15, 224–227.
40. Sakuma T, Zhao Y, Sugita M, Sagawa M, Hida M, Toga H: A prostacyclin analogue, OP-41483α-CD, restores the

ability of a β_2-adrenergic agonist to stimulate alveolar fluid clearance in rats. Surg Today, 2004, 34, 429–436.
41. Saltin B, Calbet JA: Point: in health and in a normoxic environment, VO_2 max is limited primarily by cardiac output and locomotor muscle blood flow. J Appl Physiol, 2006, 100, 744–745.
42. Sargeant AJ: Neuromuscular determinants of human performance. In: The Physiological determinants of exercise tolerance in humans. Eds. Sargeant AJ, Whipp BJ, Portland Press, London, 1999, 13–28.
43. Stergioulas AT, Filippou DK: Effects of physical conditioning on lipids and arachidonic acid metabolites in untrained boys: a longitudinal study. Appl Physiol Nutr Metab, 2006, 31, 432–441.
44. Tabet JY, Tabet JY, Meurin P, Beauvais F, Weber H, Renaud N, Thabut G et al.: Absence of exercise capacity improvement after exercise training program: a strong prognostic factor in patients with chronic heart failure. Circ Heart Fail, 2008, 1, 220–226.
45. Vanhoutte PM: Ageing and endothelial dysfunction. Eur Heart J, 2002, 4, A8–A17.
46. Wax D, Garofano R, Barst RJ: Effects of long-term infusion of prostacyclin on exercise performance in patients with primary pulmonary hypertension. Chest, 1999, 116, 914–920.
47. Wennmalm A, Nowak J, Bjuro T: Excretion of thromboxane A2 and prostacyclin metabolites before and after exercise testing in patients with and without signs of ischemic heart disease. Circulation, 1990, 82, 1737–1743.
48. Wensel R, Opitz CF, Ewert R, Bruch L, Kleber FX: Effects of iloprost inhalation on exercise capacity and ventilatory efficiency in patients with primary pulmonary hypertension. Circulation, 2000, 101, 2388–2392.
49. Whipp BJ: Domains of aerobic function and their limiting parameters. In: The Physiology and Pathophysiology of Exercise Tolerance. Eds. Steinacker JM, Ward SA, Plenum Press, New York, 1996, 83–89.
50. Womack CJ, Davis SE, Blumer JL, Barrett E, Weltman AL, Gaesser GA: Slow component of O_2 uptake during heavy exercise: adaptation to endurance training. J Appl Physiol, 1995, 79, 838–845.
51. Woodman CR, Thompson MA, Turk JR, Laughlin MH: Endurance exercise training improves endothelium-dependent relaxation in brachial arteries from hypercholesterolemic male pigs. J Appl Physiol, 2005, 99, 1412–1421.
52. Zoladz JA, Duda K, Majerczak J: Oxygen uptake does not increase linearly at high power outputs during incremental exercise test in humans. Eur J Appl Physiol, 1998, 77, 445–451.
53. Zoladz JA, Majerczak J, Duda K, Chlopicki S: Exercise-induced prostacyclin release positively correlates with VO_{2max} in young healthy men. Physiol Res, 2009, 58, 229–238.
54. Zoladz JA, Rademaker ACHJ, Sargeant AJ: Non-linear relationship between O_2 uptake and power output at high intensities of exercise in humans. J Physiol, 1995, 488, 211–217.

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