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## 1 **Introduction**

2 The questions that endurance coaches and athletes daily ask themselves are for how long, how  
3 hard and how often. In the studies by Tonnessen et al. [41, 42] it has been found that elite athletes  
4 divide their endurance training quite uniformly as 80 % of low intensity and 20 % of high intensity  
5 training. Recent research further suggests that division should be done in a polarized model [24, 40]  
6 It has been shown that both low and high intensity endurance training are needed to gain favourable  
7 peripheral adaptations in the muscle and central adaptations in the circulatory system [39]. Despite  
8 consensus according to training intensity distribution, optimal periodization during shorter  
9 microcycles and during the whole year allows much more room for different interpretations.

10 Block periodization of high intensity intervals has been shown to be an effective way to improve  
11 endurance performance [32]. There have been shorter shock microcycles lasting 1-2 weeks with  
12 almost daily HIT-sessions [2, 4] and longer periods of 4-12 weeks alternating LIT and HIT-blocks  
13 [32, 34]. Both models have improved  $VO_{2max}$ , time to exhaustion and submaximal endurance  
14 measured as speed or power at lactate threshold. It has been speculated that a high amount of highly  
15 concentrated workloads may allow greater improvements than more concurrent kind of training.  
16 The idea behind block training is to train different target abilities in series, not concurrently [18].  
17 However, actual mechanisms behind block periodization and its effects on heart rate variability  
18 (HRV), serum hormone concentrations and neuromuscular performance have remained mostly  
19 unsolved.

20 During intensive training periods that may lead to overreaching, the role of monitoring performance  
21 and fatigue becomes more important to ensure sufficient recovery. [29]Monitoring can be divided in  
22 to the external and internal methods. The external methods include performance tests like  
23 countermovement jumps [5] or submaximal performance tests [47]. The internal methods in turn  
24 include markers such as hormone concentrations of testosterone [6] or testosterone-cortisol -ratio  
25 [13] and heart rate or HRV measurements [19, 45]. HRV as a non-invasive method to evaluate the  
26 autonomic nervous system function is a potential tool to analyse a current recovery status during  
27 intensive training periods. It has been shown that HRV decreases after heavy and moderate  
28 endurance sessions [16]. Both the intensity [25, 38] and the duration of the work performed [25]  
29 may have an effect on the magnitude of delay observed in the recovery of the autonomic nervous  
30 system.

31 HRV-guided training has been studied by e.g. Kiviniemi et al. [19] and Vesterinen et al. [45]. The  
32 idea behind HRV-guided training is to adjust training load or intensity based on the autonomic

1 nervous system status. It is assumed that the decrease in HRV indicates lowered cardiac  
2 parasympathetic modulation, which in turn may be related to the reduced level of the recovery  
3 status. [16, 38] . In studies by Vesterinen et al. [45] and Kiviniemi et al. [19], HRV has been  
4 monitored daily and the intensity of daily endurance sessions have been estimated by the result of  
5 the HRV test, which further has been compared to an individually scaled reference or control value.  
6 For the definition of changes in HRV, it has been recommended to use assessment of averages of  
7 longer periods instead of individual daily values because of natural day to day variation of HRV  
8 [30]

9 Due to high demands of HIT-blocks, observed also as changes in the autonomic modulation, one  
10 may speculate that HRV-guiding of these blocks may allow a more optimal outcome compared to  
11 predetermined programming. To the best of our knowledge, no research using HRV-guiding of  
12 HIT-blocks has been published.

13 The purpose of this study was to compare predetermined and HRV-guided block periodization of  
14 HIT and its effects on endurance and neuromuscular performance, HRV and serum hormone  
15 concentrations. We hypothesized that HRV-guided training provides greater adaptations compared  
16 to predetermined training.

17

## 18 **Materials and methods**

### 19 Subjects

20 Thirty-two recreationally trained males were recruited for this study. Subjects were 19-37 years old  
21 and used to regular endurance training. Resting ECG was checked by a cardiologist before inclusion  
22 to the study. During the intervention there were 5 drop outs due to illness (n=1), injuries (n=2) and  
23 personal reasons (n=3). Three subjects were excluded due to too low adherence of training (less  
24 than 90 % of sessions). Finally, twenty-four subjects were included in the study analyses. After the  
25 control tests subjects were divided into pairs based on their age, training background, 3000 m  
26 performance and resting HRV. After that, subjects were randomly assigned to the HRV-guided  
27 group (HRV, n=13, age:  $29 \pm 4$  years, height:  $180 \pm 7$  cm, weight:  $76.4 \pm 9.4$  kg) and predetermined  
28 group (PD, n=11, age:  $31 \pm 5$  years, height:  $176 \pm 5$  cm, weight:  $74 \pm 5.7$  kg). The study was  
29 approved by the Ethics Committee of the University of Jyväskylä and it meets the ethical standards  
30 of the journal [10].

### 31 Experimental design

1 The study consisted of the 3-week control period and the 8-week training period. During the control  
2 period subjects maintained their regular amount of endurance training. However, they were  
3 instructed to plan their training so that they are fully recovered at the beginning of the training  
4 period. In addition, one interval session (3x10x30s) and one strength session was preprogrammed to  
5 make subjects familiar with these sessions before the training intervention. The control period  
6 started from the control tests and ended before the pre-tests. After the pre-tests three next days were  
7 preprogrammed in both groups, but since that the groups utilized their own training program. After  
8 four weeks of training the mid-tests were performed and the training program started from the  
9 beginning in both groups. The post-tests were performed after the 8-week training period.

10 Anthropometrics, neuromuscular measurements and 3000 m running tests were performed at the  
11 beginning of the control period, beginning of the training period, at the middle and after the training  
12 period. The tests were performed during one day so that subjects came at a fasted state to give blood  
13 samples and to perform anthropometric measurements. After these measurements a light breakfast  
14 was taken. Thereafter, the maximum running velocity test, countermovement jump (CMJ) and 1RM  
15 dynamic leg press were performed. In the afternoon the 3000 m running test was performed. The  
16 incremental treadmill test was performed before and after the training period. All the tests were  
17 performed individually at the same time of the day ( $\pm 2$  hours). Before each test at least three days  
18 of low intensity training was performed.

### 19 Training

20 Endurance training consisted of low intensity training (LIT) and high intensity training (HIT). All  
21 sessions were instructed to be performed in a flat, solid environment and individually at the same  
22 time of the day. Subjects kept Garmin XT920-heart rate monitors (Garmin Ltd, Schaffhausen,  
23 Switzerland) in each training session. They also kept training diary and wrote down a training  
24 mode, session length, heart rate and their own comments. GPS- and heart rate data of each training  
25 session was sent to the research-group to be checked manually. Every week at least one voluntary  
26 supervised session was held. From the training data a weekly training frequency, amount of  
27 endurance, other and total training and intensity distribution with the time in zone -method in a 3-  
28 zone scale (1 < 82 % HRmax, 2 = 82-87 % HRmax and 3 > 87 % HRmax) was analyzed. In  
29 addition, weekly training distribution based on the session goal of the training mode (HIT, LIT,  
30 other) was analyzed.

31 LIT-sessions were performed under the individual aerobic threshold. Subjects were instructed to  
32 maintain their typical length of LIT.sessions, but at least 30 min and at the maximum of 90 min.

1 One longer LIT-session (over 60 min) was performed every other week. Sessions included mainly  
2 running, but also alternative forms were allowed to avoid overuse injuries [45]. Two types of  
3 interval sessions were performed during the training period, which were 4x4 min intervals at the  
4 intensity corresponding to 90-95 % of maximal heart rate, individually over the anaerobic threshold  
5 with 3 min of active recovery between intervals [12] and 3x10x30 s at a running velocity equal to  
6 95 % of  $V_{\max}$  with 15 s of active recovery between intervals and 3 min between sets [33].

7 A strength session was performed five times during the training period. The session was a mixture  
8 of maximum and explosive strength training. Leg press, knee flexion and two upper body exercises  
9 were performed at loads of 70-85 % of 1RM with 2-3 sets and 5-10 repetitions. Bench step (body  
10 weight) and half squat (30/40/60 %/1RM) were performed as explosively as possible using 3 sets of  
11 5-6 repetitions. In addition, two core exercises were performed with three sets of 20 repetitions.

12 The training at the PD group consisted of HIT-block weeks (4-5 HIT-sessions) and recovery weeks  
13 (1 HIT-session) which were performed in turns through the training period [34]. One rest day was  
14 included in each week. The same training program model was used also during the second period  
15 (weeks 5-8) (Figure 1).

16 The HRV group had the same training modes as the PD group, but the way of programming those  
17 modes differed. In HRV, the training program was divided into six blocks (Figure 1). Each block  
18 was preprogrammed, but moving from block to another took place based on the quick recovery test  
19 result (Firstbeat technologies Ltd, Jyväskylä, Finland) performed each morning. The control period  
20 allowed the software to adapt to the individual range of heart rate and HRV. During the training  
21 period a 3-day running average of the quick recovery test results was used for the training guidance  
22 due to day to day variation in HRV [30]. Individual average of all the quick recovery test results  
23 from the control period was used as a reference value for starting the next block. After finishing the  
24 predetermined block, LIT was performed until the 3-day running average of the QRT score was  
25 higher than the individual reference value. After the mid-tests or finishing block 6, subjects started  
26 the training program from block 1. The strength sessions were programmed by the research group  
27 between HIT blocks, so that they were performed as a LIT-session and at the same time as in the  
28 PD group.

29 Subjects performed the quick recovery test (Firstbeat technologies Ltd, Jyväskylä, Finland) every  
30 morning during the control and the training period. The test required a 3 min of RR-interval data  
31 collection. The software performed artefact correction and data filtering [35]. The RR-interval data  
32 was collected with Garmin 920XT heart rate monitor (Garmin Ltd, Schaffhausen, Switzerland). The

1 quick recovery test score was derived by the Firstbeat SPORTS Monitor v. 2.0 (Firstbeat  
2 Technologies Ltd, Jyväskylä, Finland) using heart rate and RMSSD parameters for describing vagal  
3 activity. The results were adaptively scaled based on the average and standard deviation of the user's  
4 personal measurement history. The results were presented from 0 to 100% (0-30% poor, 30-70%  
5 moderate, 70-90% good, and 90-100% excellent recovery).

### 6 3000 meter running and incremental treadmill tests

7 The 3000 m running test was performed in the 200 m indoor running track. Before the test a 15 min  
8 warm up was performed. The running tests were performed in groups of average 4 subjects. After  
9 the test lactate samples were taken from the fingertip immediately after and 4 min after the end of  
10 the test (Biosen S\_line Lab+ lactate analyzer, EKF Diagnostic, Magdeburg, Germany). In addition,  
11 average heart rate and maximum heart rate were analyzed.

12 The incremental treadmill test was performed in the laboratory of Biology of Physical Activity at  
13 the University of Jyväskylä (Telineyhtymä, Kotka, Finland). The test started at the velocity of 8  
14 km/h or 10 km/h based on the fitness level of each subject. The same starting velocity was used in  
15 both tests. The 8 km/h velocity was increased to 10 km/h and by 1 km/h at each 3 min stage  
16 thereafter. After each stage the treadmill was stopped for fingertip blood samples (20 s). Lactate  
17 samples were analyzed with Biosen S\_line Lab+ lactate analyzer (EKF Diagnostic, Magdeburg,  
18 Germany). During the test heart rate was recorded with Garmin XT920 -heart rate monitor (Garmin  
19 Ltd, Schaffhausen, Switzerland). The incline was kept at 0.5 degrees through the test. Oxygen  
20 consumption was measured through the test breath by breath. (OxyconPro, Jaeger, Hoechberg,  
21 Germany). Before each test the gaz analyzer was manually calibrated.

22  $VO_{2max}$  was defined as the highest 60 s average of oxygen consumption.  $V_{max}$  was defined as the  
23 highest speed finished, or if the stage was not finished, as a speed of the last completed stage (km/h)  
24 + running time of the unfinished stage (s)-30 s/180 s\*1 km/h. (running time (s) of the speed at  
25 exhaustion - 30 s) / 180-30 s) Aerobic (LT1) and anaerobic (LT2) thresholds were determined  
26 based on lactate values during the test. The aerobic threshold was set at 0.3 mmol/l above the lowest  
27 lactate value and the anaerobic threshold at the intersection point between 1) a linear model  
28 between LT1 and the next lactate point and 2) a linear model for the lactate points with the La  
29 increase of (at least) 0.8 mmol/l. [46]

### 30 Anthropometrics and neuromuscular measurements

1 Body weight and fat percentage was analyzed after 12 hours of fastning with InBody720-analyzer.  
2 (InBody720 body composition analyzer, Biospace Co. Ltd, Seoul, South-Corea).

3 The maximum running velocity test was performed in the indoor track. Before the test subjects  
4 performed 10 min warm up, dynamic streching and three accelerations of 40-50 meters. The  
5 maximum velocity (m/s) was calculated from the 10 m running distance between the photocells  
6 after the 25 m acceleration. Subjects had three attempts unless more than 5 % improvement was  
7 observed between the second and third attempts. Between the attempts a recovery of 2 min was  
8 allowed.

9 Countermovement jumps were performed on the force plate (Departmenet of Biology of Physical  
10 Activity, Jyväskylä, Finland). During the jumps subjects held hands on their hips. The starting  
11 position was instructed to be at 90 degrees knee angle. Three jumps were performed with 1 min  
12 recovery between jumps, unless more than 5 % improvement was observed between the second and  
13 third attempts. Jumping height was analyzed from the force impulse. The analysis was done with  
14 the Signal 4.10 -program (Cambridge Electronic Design Ltd, Cambridge, UK).

15 The dynamic leg press action was performed concentrically using the David 210 dynamometer  
16 (David Sports Ltd., Helsinki, Finland). The starting knee angle was individually set to 60 degrees.  
17 The warm up protocol consisted five repetitions with the loads at 70 % of 1RM, three repetitions at  
18 80 % of 1RM and two repetitions at 90 % of 1RM. Between the sets one-minute recovery was  
19 allowed. After the warm up, one repetition at a time was performed until the subject could not finish  
20 the increased load. Between the repetitions a 1.5 min recovery was allowed.

## 21 Heart rate variability

22 Heart rate and HRV was recorded every morning and every other night through the study period.  
23 The morning measurements were done with Garmin 920XT heart rate monitors (Garmin Ltd,  
24 Schaffhausen, Switzerland). The measurement was instructed to be performed right after awakening  
25 and emptying the urinary bladder. The measurement was a 3 min long and it was performed in a  
26 supine position. Before starting the data collection, subjects were instructed to wait until heart rate  
27 became steady. Subjects sent HR-data to the research-group and data was analyzed with the  
28 Firstbeat Sports software. Weekly average of the morning heart rate and RMSSD was analyzed.  
29 Nocturnal measurments were done with the Firstbeat Bodyguard device (Firstbeat Technologies  
30 Ltd., Jyväskylä, Finland). Subjects were instructed to put the device on when going to sleep and to  
31 release the device immediately after awakening. From the nocturnal measurements a 4-hour period  
32 starting 30 min after going to sleep was analyzed. Recorded RR-intervals were edited by an artifact

1 detection filter of the Firstbeat Sports software, which excluded all falsely detected, missed, and  
2 pre-mature heartbeats. If the error percentage representing the amount of corrected interbeat  
3 intervals shown by the software was higher than 33 %, recordings were excluded from the analysis  
4 in line with the suggestion by Vesterinen et al. [44]. Heart rate, RMSSD, low frequency (LF), high  
5 frequency (HF), and total power (TP) were analyzed from the whole control period and during the  
6 training weeks 4 and 8.

### 7 Serum hormone concentrations

8 Serum hormone concentrations were measured at the same time of the day (8:00-9:00) and after 12  
9 hours of fasting. Blood samples were taken from the antecubital vein into serum tubes  
10 (Vacuette, Greiner Bio One International GmbH, Bad Haller Str. 32 4550 Kremsmünster, Austria)  
11 using standard laboratory procedures. Whole blood was centrifuged at 3500 rpm (Megafuge 1.0 R,  
12 Heraeus, Hanau, Germany) for 10 min. After that serum was removed and fridged at -80 degrees  
13 until the final analysis. Serum testosterone and cortisol were analyzed with chemical luminescence  
14 techniques (Immulite 2000 XPi, Siemens, New York City, NY, USA) and hormone specific  
15 immunoassay kits (Siemens, New York City, NY, USA). The sensitivity of testosterone and cortisol  
16 assays were 0.5 nmol/l and 5.5 nmol/l, respectively. The intra-assay coefficients of variation for  
17 testosterone and cortisol was 7.3 % and 8.3 %, respectively.

### 18 Statistical analysis

19 All the values are presented as mean  $\pm$  standard deviation. Normality of the data was assessed with  
20 the Shapiro-Wilk test. To test for differences between the groups at baseline and within groups  
21 between the control and pre-tests unpaired two-tailed t-tests and paired two-tailed t-tests were used.  
22 Within group differences at the incremental treadmill test were compared using paired two-tailed t-  
23 test ( $VO_{2max}$ ,  $V_{max}$ , LT1, and LT2). Neuromuscular performance, 3000 m test, serum hormone  
24 concentrations and HRV were analyzed using repeated measures ANOVA. If the ANOVA reached  
25 significance, a Fisher's LSD test was performed for post hoc analysis.  $VO_{2max}$  l/min and LF ( $ms^2$ )  
26 values of the PD-group was not normally distributed so the data was analyzed with nonparametric  
27 Wilcoxon signed rank test. To test for differences in relative changes from the pre-intervention to  
28 post-intervention between the groups unpaired Students t-tests were performed. In addition, effect  
29 size (ES) of between group differences in the relative changes of key performance and  
30 physiological variables was calculated as Cohen's d. The magnitude of changes was stated as < 0.2  
31 trivial, 0.2-0.5 small, 0.5-0.8 moderate and >0.8 large. The correlation analysis was done using  
32 Pearson moment product method. Significance was set at  $p \leq 0.05^*$ ,  $p < 0.01^{**}$  and  $p < 0.001^{***}$ .



1 Results were analyzed with Microsoft Excel 2010 (Microsoft Corporation, Redmond, WA, USA)  
2 and IBM SPSS Statistics v.24 -programs (SPSS Inc, Chicago, IL, USA).

3

#### 4 **Results**

5 There were no significant between group differences in the baseline levels of endurance,  
6 neuromuscular, HRV or serum hormone concentration variables. No significant changes were found  
7 during the control period, except for maximal running velocity in HRVG.

#### 8 Anthropometrics

9 There were no significant changes from pre to post training period in body weight (HRV  $76.5 \pm 9.0$   
10 kg vs.  $76.4 \pm 9.4$  kg; PD  $74.0 \pm 5.5$  vs.  $74.0 \pm 5.7$  kg) or fat percent (HRVG  $12.6 \pm 4.2$  % vs.  $12.6 \pm$   
11  $4.4$  %; PD  $12.6 \pm 2.7$  % vs.  $12.2 \pm 3.2$  %).

#### 12 Training

13 No significant differences were observed between the groups in the amount of training or training  
14 intensity distribution during the control and training periods. In both groups significant increases  
15 were found from the control period to the training period in training frequency, and amount of Zone  
16 2 training (HRVG  $5.3 \pm 2.1$  vs.  $6.3 \pm 1.4$ ,  $p=0.007$ ; PD  $5.0 \pm 1.1$  vs.  $6.1 \pm 0.4$ ,  $p=0.001$ ) (HRVG  $10$   
17  $\pm 7$  % vs.  $15 \pm 6$  %,  $p<0.005$ ; PD  $6 \pm 4$  % vs.  $12 \pm 5$  %,  $p=0.008$ ). In the PD group a significant  
18 increase was found in the amount of endurance and total training ( $4.7 \pm 1.7$  vs.  $5.3 \pm 1.8$   $p<0.001$ )  
19 and ( $5.2 \pm 1.8$  vs.  $6.0 \pm 1.9$   $p<0.001$ ). Training characteristics of both groups during the training  
20 period are presented in table 1.

21 No significant differences were found between the groups in the number of HIT-sessions during the  
22 training period. The total number of HIT-sessions in PD and HRVG were, on average,  $21.8 \pm 0.6$   
23 vs.  $19.8 \pm 4.1$ , respectively. The PD group performed, on average,  $10.9 \pm 0.3$  HIT-sessions during  
24 the first and last four weeks of the training period, while the HRVG group performed  $10.3 \pm 2.7$   
25 HIT-sessions during the first four weeks and  $9.5 \pm 2.8$  HIT-sessions during the last four weeks. No  
26 significant correlation was found between the number of HIT-sessions and endurance performance  
27 changes in the HRVG group.

28 In the weekly training distribution significant differences were found between the groups. During  
29 weeks 2 ( $p=0.008$ ) and 7 ( $p=0.030$ ) the relative amount of weekly HIT-sessions was significantly

1 different between groups. During weeks 1,3,4,6,8 between group differences in the relative amount  
2 of HIT-sessions approached the significance level ( $p=0.054-0.075$ ) (Figure 2).

### 3 Endurance performance

4 Both groups increased their  $V_{\max}$  significantly (HRVG  $p<0.001$ ; PD  $p<0.001$ ). A significant  
5 difference and large effect size ( $ES=0.95$ ) between the groups was found in the relative increase of  
6  $V_{\max}$  ( $p=0.033$ ) (Figure 3).  $VO_{2\max}$  relative to body weight and absolutely increased in HRVG  
7 ( $p=0.001$ ,  $p=0.011$ ) and PD ( $p=0.005$ ,  $p=0.036$ ). Moderate effect size ( $ES=0.52$ ) was found between  
8 the groups in the relative increase of absolute  $VO_{2\max}$ . Anaerobic threshold (LT2) increased in both  
9 groups significantly (HRVG  $p<0.001$ ; PD  $p=0.050$ ). Significant increases were found in the aerobic  
10 threshold (LT1) in HRVG ( $p=0.021$ ) and PD ( $p=0.027$ ) (Table 2).

11 Both groups improved their performance in the 3000 m test from pre to post (HRVG  $-5.2 \pm 2.4$  %,  $p<0.001$ ;  
12 PD  $-5.2 \pm 3.1$  %,  $p=0.001$ ), pre to mid- (HRVG  $-3.1 \pm 1.3$  %,  $p<0.001$ ; PD  $-3.5 \pm 2.6$  %,  $p=0.002$ )  
13 and mid to post (HRVG  $-2.2 \pm 1.5$  %,  $p<0.001$ ; PD  $-1.5 \pm 1.1$  %,  $p=0.001$ ). Maximum  
14 lactate values increased significantly in the HRVG group from mid to post ( $12.8 \pm 18.4$  %,  $p=0.039$ ).  
15 In the PD group a same kind of trend was observed from pre to post ( $16.0 \pm 23.5$  %,  $p=0.064$ ) (Table 3).

### 17 Neuromuscular performance

18 No significant changes were found within groups in the CMJ during the training period. From pre to  
19 mid jumping height in PD tended to decrease ( $29.0 \pm 3.8$  cm vs.  $28.4 \pm 3.7$  cm,  $p=0.073$ ), while  
20 increasing trend were seen in HRVG from pre to post ( $31.4 \pm 4.8$  cm vs.  $32.1 \pm 5.2$  cm). A significant  
21 difference between the groups was found in the relative change of CMJ from pre to post ( $p=0.048$ )  
22 with large effect size ( $ES=0.88$ ) (Figure 4).

23 Maximal running velocity increased in HRVG significantly from the control to the pre test ( $8.14 \pm$   
24  $0.31$  m/s vs.  $8.20 \pm 0.32$  m/s,  $p=0.008$ ). From pre to mid test maximal running velocity in PD  
25 decreased significantly ( $7.95 \pm 0.40$  m/s vs.  $7.87 \pm 0.36$  m/s,  $p=0.008$ ) and tended to decrease in  
26 HRVG ( $-0.5 \pm 1.0$  %,  $p=0.054$ ). No other significant differences were found between or within  
27 groups (Figure 4).

28 1RM increased from pre to post significantly in both groups (HRVG;  $206 \pm 29$  kg vs.  $229 \pm 32$  kg,  
29  $p=0.001$ ; PD  $202 \pm 32$  kg vs.  $225 \pm 33$  kg,  $p<0.001$ ). From pre to mid only PD increased leg press  
30 significantly ( $1.9 \pm 3.0$  %  $p=0.024$ ). From mid to post HRVG ( $8.7 \pm 6.6$  %,  $p<0.001$ ) and PD ( $9.5 \pm$

1 6.3 %,  $p=0.001$ ) increased their 1RM. No significant differences were found between the groups in  
2 the relative change of 1RM.

### 3 Heart rate variability

4 Nocturnal heart rate decreased significantly from pre to post in both groups (HRVG;  $p=0.004$  and  
5 PD;  $p=0.008$ ) (Table 3). The decrease in HR from pre to mid was significant only in PD ( $p<0.042$ ).  
6 In HRVG a significant increase was observed from pre to post in RMSSD ( $p=0.028$ ), LF ( $p=0.024$ ),  
7 and TP ( $p=0.046$ ), while no significant changes were found in PD. In the morning measurements no  
8 significant differences were observed in the relative change of RMSSD from the control period to  
9 any week during the training period (Figure 5). Heart rate was significantly lower compared to the  
10 control period during weeks 4-8 ( $p<0.05$ ) in HRVG and during 3,7 and 8 in PD ( $p<0.05$ ). Effect  
11 size showed moderate between group effect in heart rate change from the control period to week 8  
12 ( $ES=0.65$ ) and small effect in RMSSD ( $ES=0.42$ )

### 13 Serum hormone concentrations

14 Serum testosterone concentration decreased significantly in PD from pre to mid ( $p<0.037$ ), while no  
15 significant change was observed in HRVG (Table 3). From mid to post testosterone increased  
16 significantly in HRVG ( $p<0.029$ ). Effect size showed moderate between group effect in  
17 testosterone change from pre to post. No significant within or between group differences were  
18 found in the serum concentration of cortisol or the testosterone/cortisol-ratio, although  
19 testosterone/cortisol-ratio increase approached significance from mid to post in HRVG ( $p=0.051$ ).

### 20 Correlations

21 A significant correlation was found between individual baseline HF and individual changes in  $V_{max}$   
22 in PD ( $r=0.656$ ,  $p=0.028$ ) (figure 6), while no such a correlation was observed in HRVG. Individual  
23 resting HR changes from pre to post correlated with 3000 m changes ( $r=-0.630$ ,  $p=0.01$ ), as well as  
24 individual HF ( $r=0.488$ ,  $p=0.018$ ) and TP ( $r=0.467$ ,  $p=0.025$ ) changes from pre to post in the total  
25 group of subjects. In the morning measurements individual RMSSD changes from the control  
26 period to the weeks 5-8 correlated significantly with  $V_{max}$  changes ( $r=0.499$ ,  $p<0.015$ ) in all  
27 subjects. In the nocturnal measurements individual changes from mid to post in HF ( $r=0.414$ ,  
28  $p=0.049$ ) and TP ( $r=0.485$ ,  $p=0.019$ ) correlated with  $V_{max}$  changes in the total group of subjects.  
29 Individual TP ( $r=0.462$ ,  $p=0.026$ ) and HF ( $r=0.503$ ,  $p=0.014$ ) changes from pre to post correlated  
30 positively with absolute average serum testosterone concentrations and average  
31 testosterone/cortisol-ratio ( $r=0.479$ ,  $p=0.021$  and  $r=0.465$ ,  $p=0.025$ , respectively) in all subjects. In

1 HRVG a significant correlation was found between absolute morning RMSSD values and the  
2 number of HIT-sessions during the last four weeks ( $r=0.592$ ,  $p<0.05$ ).

3 Individual average serum testosterone concentrations (pre, mid and post) correlated significantly  
4 with individual changes in  $V_{\max}$  ( $r=0.510$ ,  $p=0.01$ ) (figure 6) and in 3000 m ( $r=0.570$ ,  $p<0.01$ ) in all  
5 subjects. A significant correlation was also found between average serum testosterone/cortisol-ratio  
6 and changes in  $V_{\max}$  ( $r=0.457$ ,  $p=0.025$ ) and 3000 m ( $r=0.510$ ,  $p=0.011$ ) in the total group of  
7 subjects. Individual changes in testosterone concentrations from mid to post correlated positively  
8 with changes in  $V_{\max}$  ( $r=0.527$ ,  $p<0.01$ ) in all subjects. Individual changes in CMJ from mid to post  
9 correlated positively with changes in  $V_{\max}$  ( $r=0.469$ ,  $p=0.021$ ) in the total group of subjects.

## 10 **Discussion**

11 Both groups improved significantly 3000 m running result and endurance performance in the  
12 incremental treadmill test. However, the main finding of the current study was the significantly  
13 greater increase in  $V_{\max}$  and countermovement jump after HRV-guided compared to pre-determined  
14 training after 8-weeks of High-Intensity block training. Significant increases in HRV and serum  
15 testosterone concentration were observed in HRVG, but not in PD. This study suggests that  
16 individually HRV guided programming of HIT blocks contributes to greater positive adaptations  
17 compared to predetermined training.

## 18 Training

19 Training intensity distribution as time in the zone approach was almost identical between the  
20 groups. When analyzing weekly distribution with the session goal approach, significant differences  
21 were observed. In HRVG, blocks tended to be performed in a more even way through the training  
22 period. In previous HRV-guiding studies significant differences between predetermined and HRV-  
23 guided groups have been observed in the amount of HIT-training [19, 45]. Although no significant  
24 differences were observed in the present study, much larger interindividual variation in the amount  
25 of HIT-sessions was found in HRVG ( $SD=4.1$ ) compared to PD ( $SD=0.6$ ). It seems that some  
26 individuals were able to recover and benefit to much a greater amount of HIT sessions than others.  
27 During short training periods of 1-2 weeks HIT-sessions have been performed almost daily, but not  
28 in longer interventions [2,4]. No significant correlation was found between the individual amount of  
29 HIT-sessions and endurance performance adaptations in HRVG, suggesting that similar adaptations  
30 can be gained with differently periodized HIT training.

## 31 Endurance performance

1 Both groups improved endurance performance in the incremental treadmill test and in 3000 m  
2 running. Also  $VO_{2max}$  and velocity at the thresholds increased significantly in both groups. The  
3 magnitudes of improvements were in line with previous block periodization [32] and HRV guiding  
4 studies [19, 45]. Every subject improved their  $V_{max}$  on the treadmill and also running time in 3000  
5 m. Despite that, a significant difference and large between group effect size was found in the  
6 relative change of  $V_{max}$ . In addition, moderate between group effect size was noted in the change of  
7 absolute  $VO_{2max}$ . These findings were interesting due to the almost identical improvement of 3000  
8 m in both groups. They were also somewhat different compared to the study by Vesterinen et al.  
9 [45] where the HRV-guided group performed superior only in 3000 m, but not on the treadmill.  
10 This difference might be explained via different kinds of periodization of the training in the  
11 predetermined group, as in the current study it was more similar to the HRV-guided group.  
12 Neuromuscular performance and fatigue may at least partly explain the observed group difference  
13 in  $V_{max}$ , because the significant between group difference was found also in the CMJ change. No  
14 correlation was found between individual pre-post changes in CMJ, but mid-post individual  
15 changes in CMJ correlated significantly with individual relative changes of  $V_{max}$ . The negative trend  
16 in CMJ may indicate neuromuscular fatigue caused by too high an amount or badly timed HIT-  
17 blocks.  $V_{max}$  speed was on average 10 % higher than average speed during the 3000 m test,  
18 indicating that neuromuscular demand at  $V_{max}$  may be higher. As Paavolainen et al. [28] stated a so  
19 called muscle power factor may be an important determinant of maximal running performance.

## 20 Neuromuscular performance

21 To the best of our knowledge, no previous study has investigated the effects of block periodization  
22 of HIT-sessions on neuromuscular performance. In the current study the significant between group  
23 difference with large effect size was found in the CMJ change. While CMJ increased in HRVG, it  
24 decreased in PD. This finding is in line with the Vesterinen study [46], where only the HRV-guided  
25 group improved reactivity jump during the training period. A too high amount of endurance training  
26 may disturb neuromuscular adaptations and performance, especially rapid force development [17,  
27 23] which was observed also as decreased maximal running velocity during the first four weeks of  
28 the present training intervention. In previous running interventions both aerobic and supramaximal  
29 intervals have improved maximal running velocity and endurance performance [3]. It seems that  
30 block periodization of HIT may be more challenging in the perspective of rapid force development.  
31 Gomez et al. [7] found decreased force production in isokinetic knee flexion and CMJ after the 10  
32 kilometer race still 48 hours after the race. Demands of the interval sessions used in the present  
33 study may not be as high, but a similar type of trend might be possible. During HIT-blocks of the

1 current study such a long recovery time was not allowed, and it can be speculated that heavy HIT  
2 microcycles with insufficient recovery, may lead to cumulated fatigue and the decrease in  
3 neuromuscular performance. HRV-guiding seems to allow more optimal recovery between blocks.  
4 Despite between group differences observed in CMJ, both groups increased significantly 1RM leg  
5 press. This improvement can be partly explained via learning effects, but it seems, that already a  
6 very low amount of strength sessions as used in this study combined with HIT-training may be  
7 enough to gain some measurable adaptations in maximum force, at least, in not strength trained  
8 subjects.

9 According to neuromuscular performance, also running biomechanics need to be considered. While  
10 cycling, for example, includes mostly concentric muscle action, running involves stretch-shortening  
11 cycles with an eccentric component [20]. Microdamage in muscle may decrease endurance  
12 performance and that way lead to a decreased training stimulus during intensive periods [1, 22].  
13 Earlier block periodization studies have not included runners as subjects [2, 4, 32, 34]. This aspect  
14 is important to notice when planning block training for runners.

#### 15 Heart rate variability

16 Nocturnal HRV increased in the HRVG group, while only the positive trend was observed in PD.  
17 Earlier investigations have found no significant change [43], an increase [27] and an acute decrease  
18 [31] in HRV following intensive training. In the current study, both the absolute values and the  
19 increase of parasympathetic HRV markers (nocturnal HF and TP, morning RMSSD) were  
20 associated with positive adaptations to endurance training, which strengthens the use of HRV in  
21 monitoring endurance training load and adaptations. In addition to HRV the significant decrease in  
22 nocturnal heart rate was observed similarly to previous studies [36, 44]. In the current study the  
23 magnitude of decrease was quite great considering the short amount of time and the subjects who  
24 already had low baseline values. This finding may suggest that specific cardiac adaptations such as  
25 increased stroke volume was caused by block periodization.

26 Despite that in the current study increased HRV was associated with improved performance, it has  
27 not been the case in all other studies. For example, in the study by Le Meur et al. [21] significant  
28 parasympathetic hyperactivation was found followed by overreaching. Overreaching was achieved  
29 with the increase of the training volume so it can be speculated that reactions may differ between  
30 training interventions with increased training volume or intensity. Schmitt et al. [37] also underlined  
31 the individuality of HRV reactions followed by intensive training. They found four different kinds

1 of fatigue shifts of HRV patterns. More research is still needed to examine different types of  
2 individual HRV reactions and how they are possibly related to the type of training performed.

3 An interesting, but not a novel finding was the association between baseline HF and  $V_{\max}$  change in  
4 PD. [11, 44] In the Vesterinen et al. [44] study a same kind of correlation was found between the  
5 individual baseline HF and  $V_{\max}$  adaptations to HIT-training. The correlation was negative with  
6 LIT-training. Based on the association found in PD, but not in HRVG, it might be more about the  
7 timing and amount of HIT-training than about the intensity or volume of the training. In the present  
8 study, the significant correlation was found between the individual morning RMSSD during the last  
9 four weeks of the training period and the number of hit-sessions in HRVG. This also may be related  
10 to the link between absolute HRV and capability to cope with high amounts of high intensity  
11 training. Based on the correlations found between the individual HRV mid to post changes and  
12 individual changes in performance, these associations may come more critical as the length of the  
13 intensive training period increases.

14 Morning and nocturnal measurements of HRV showed slightly different trends as has been found  
15 also in previous studies. The morning measurements were used for periodization in the current  
16 study because of practical reasons. Despite that the nocturnal measurements are often stated to be a  
17 more standardized method, in Hynynen et al. [15] study no changes in nocturnal HRV markers were  
18 observed in the overtrained athletes, while in the morning measurements significant decreases were  
19 found. The authors speculated that waking up causes always a kind of stress reaction which may  
20 lead to different results compared to the night measurement. In the present study there were no  
21 significant differences between the groups according to nocturnal heart rate or HRV changes during  
22 the training period. In the morning measurements the small between group effect size in RMSSD  
23 and moderate in heart rate was observed when comparing the relative change from the control  
24 period to the week-8 values.

25 Due to different results obtained from different kinds of measurements, it is important to use always  
26 the same kind of protocol. In addition, using averages instead of individual values is highly  
27 recommended as stated by Plews et al. [30]. In the current study the 3-day rolling average of the  
28 quick recovery test was used. Previous studies have used 7-day averages [45] and daily values [19].  
29 Averaging results for a longer period may decrease the risk of false results due to high day to day  
30 variation in HRV [30], but at the same time averages of a very long period may make it difficult to  
31 react fast for changes in the autonomic nervous system. Average of three-four days may be a good  
32 compromise, as it decreases the value of an individual result, but still makes it possible to react fast

1 to observed trends. The reference value plays also an important role as regulating the start of HIT-  
2 blocks. In the current study the average value of the control period was used, which seemed to  
3 allow good recovery for most of the subjects. Few individuals had troubles to obtain their test result  
4 over the reference value, probably due to stress outside the training. Despite a low amount of HIT  
5 blocks, they still improved their performance. The quick recovery test scaled the result based on  
6 individual measurement history. In that way the reference value was continuously updated as more  
7 HRV data was collected. The updating reference value and SWC during longer training periods  
8 may be recommended as significant increases of HRV markers were observed in the current study  
9 followed by training.

#### 10 Serum hormone concentrations

11 It has been found that endurance athletes tend to have lower testosterone concentrations compared  
12 to controls [8]. However, adaptations observed after the endurance training period has varied from  
13 the decrease [13], increase [6] to no change [43]. Training mode may affect on hormonal response,  
14 since greater acute free testosterone response has been found after high intensity training session  
15 compared to low intensity training session [8]. In the current study the significant increase in serum  
16 testosterone concentration and the tendency of increase in the testosterone/cortisol-ratio was  
17 observed from mid to post in the HRVG group. The significant decrease was noted in testosterone  
18 in PD from pre to mid. No significant changes were observed in concentrations of other hormones  
19 examined.

20 Individual basal serum testosterone concentration as well as the testosterone-cortisol ratio correlated  
21 with changes in  $V_{\max}$  and 3000 m. Hoogeveen and Zonderland [13] found no correlation between  
22 the improvement of cycling performance and changes in testosterone or cortisol during a training  
23 period. However, Mäestu et al. [26] concluded that the first sign of decreased adaptivity in athletes  
24 is a decreased resting level of free testosterone and a lower maximal exercise-induced acute  
25 increase in free testosterone concentration. Most studies have focused on typical high volume  
26 endurance training, so it can be speculated that high intensity training may induce different  
27 adaptations. Zinner et al. [47] found that after 2 weeks of HIT training a positive correlation  
28 between the improvement in endurance performance and an increase in basal testosterone  
29 concentration was observed.

30 An interesting relationship was also found between individual absolute testosterone concentrations  
31 and testosterone/cortisol-ratios and individual changes of HRV during the present training period.  
32 Similar to that Huovinen et al [14] found a significant correlation between the testosterone/cortisol-



1 ratios and the changes in HF during the stressful first week of military service. According to  
2 intensive block training, in addition to positive changes in testosterone and the testosterone-cortisol-  
3 ratio, also higher absolute serum testosterone concentrations may be beneficial.

#### 4 Conclusions

5 The present results suggest that block periodization of HIT is an effective way to improve  
6 endurance and running performance in a short amount of time in already endurance trained males.  
7 Individually HRV guided timing and the amount of HIT-blocks seems to provide greater endurance  
8 and neuromuscular adaptations compared to predetermined training. Individually guided training  
9 may to reduce risk of overtraining observed as positive changes in HRV and serum testosterone  
10 concentrations. Both baseline heart rate variability and testosterone levels may to be associated  
11 with the capacity of an individual to adapt to intensive block training.

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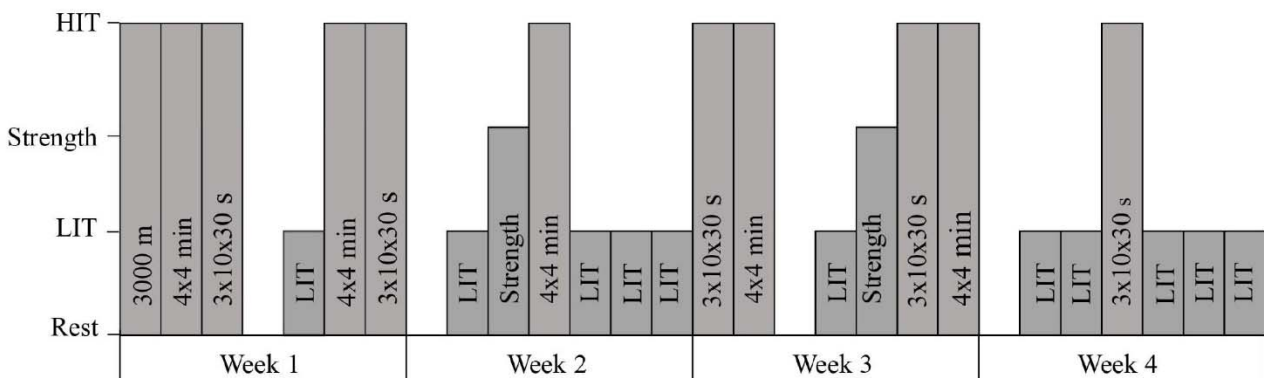
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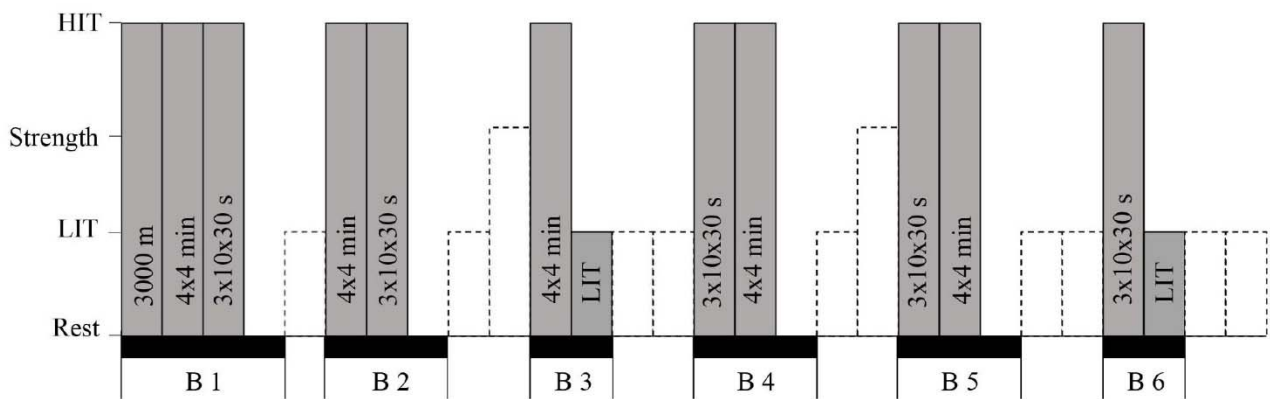
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1 **Figure legends**

a) Training program in the PD group



b) Training program in the HRVG group



2

3 **Figure 1.** Description of the training program in PD and HRVG. In PD the program was divided to  
 4 HIT-block weeks and recovery weeks. HRVG had the same training modes, but the program was  
 5 divided to six blocks (B1-B6). Moving from block to another took place based on the quick  
 6 recovery test result. Only LIT-sessions were performed in HRVG as long as the test result was  
 7 below the individual reference values. Strength sessions were performed as LIT-sessions and they  
 8 were placed by research group. Both groups started the similar training program from the beginning  
 9 after four weeks of training.

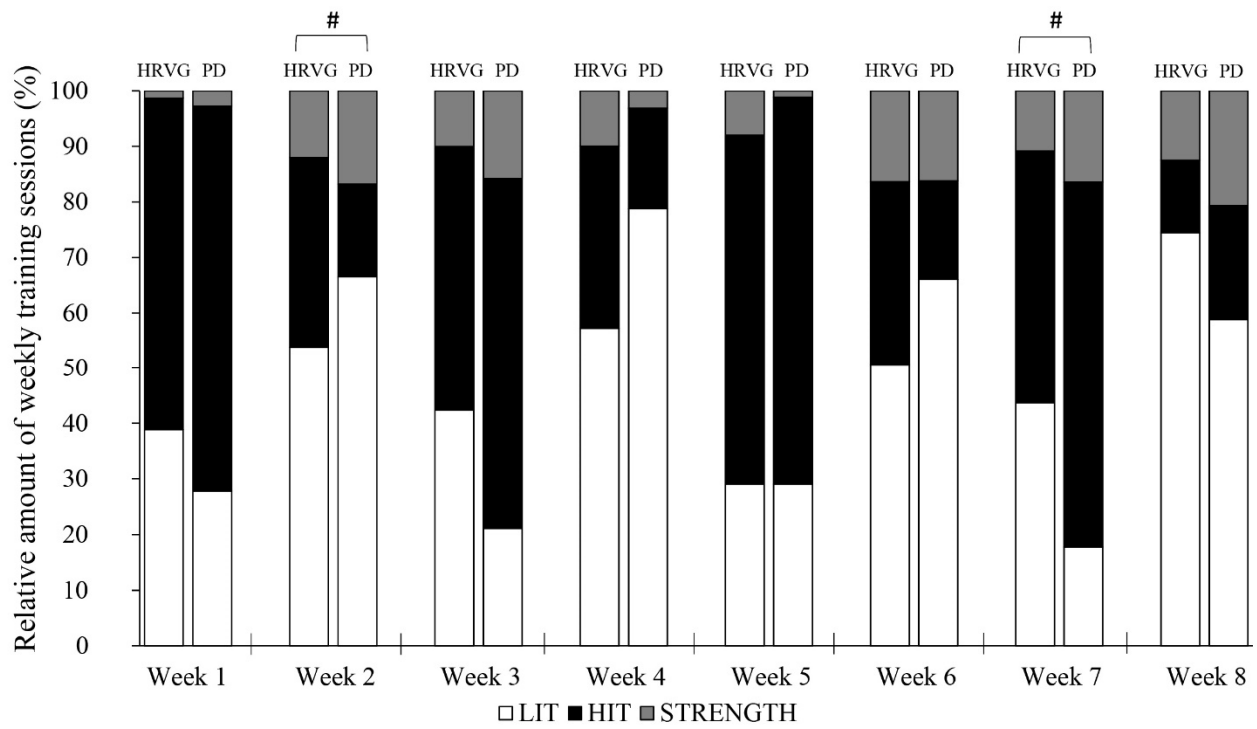
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2 **Figure 2.** Weekly average distribution of different training sessions in both groups. LIT = low  
 3 intensity training, HIT = high intensity training, strength = strength training. Statistical  
 4 significances: # p<0.05, ## p<0.01, between groups difference

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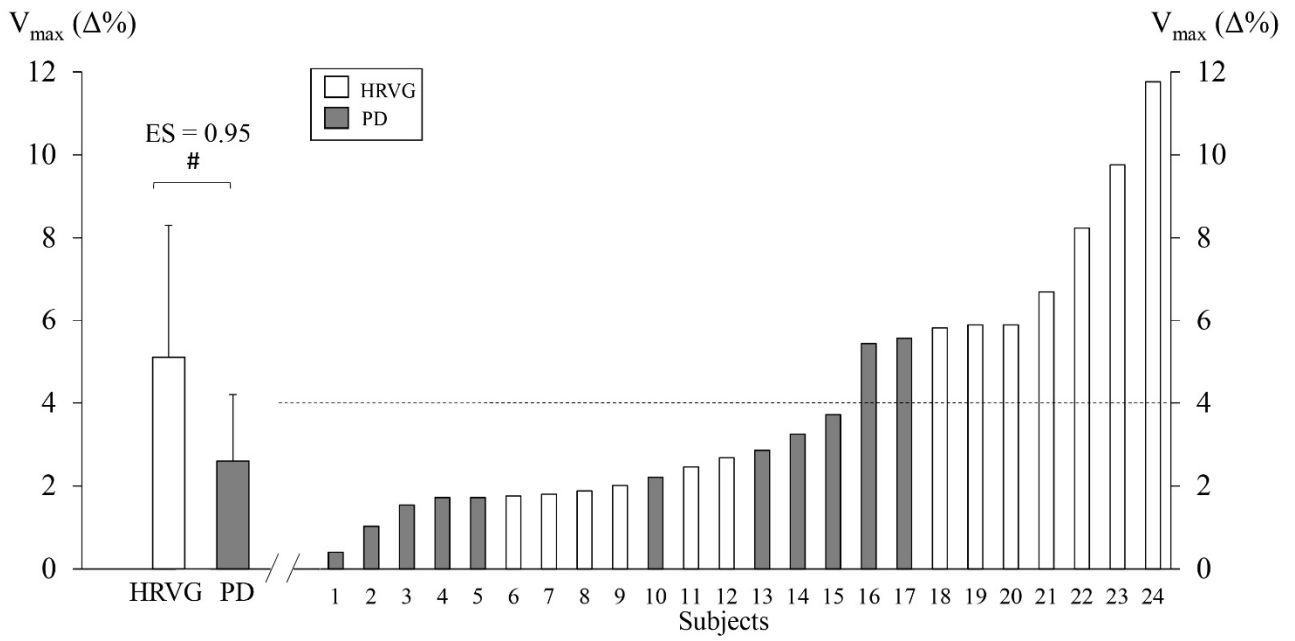
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2 **Figure 3.** Relative mean changes of the groups and individual changes of each subject in  $V_{max}$ .  
 3 Black line presents the mean of the whole group (4.0 %). Statistical significances: #  $p < 0.05$   
 4 between groups difference

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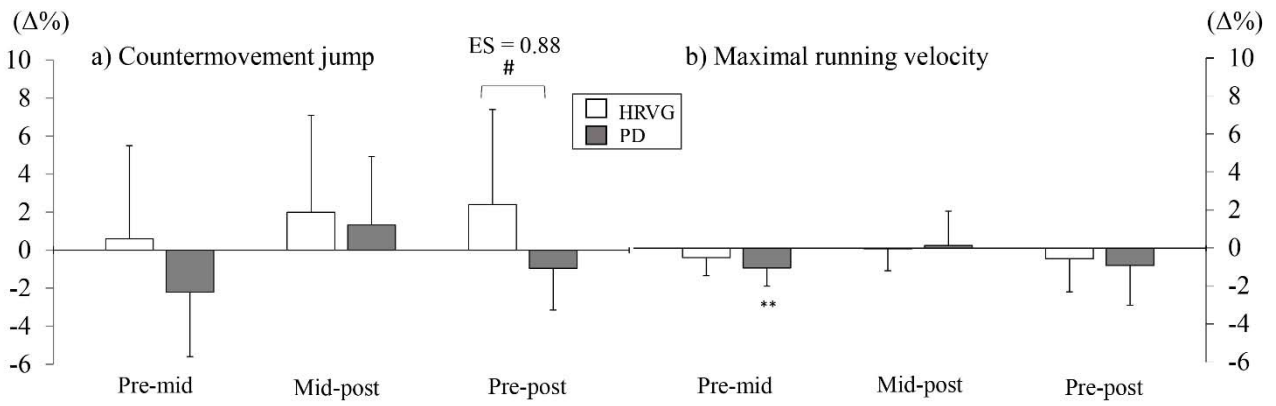
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2 **Figure 4.** Relative changes in countermovement jump and maximal running velocity from pre-mid,  
 3 mid-post and pre-post. Statistical significances: \*\*  $p < 0.01$  within groups, #  $p < 0.05$  between groups

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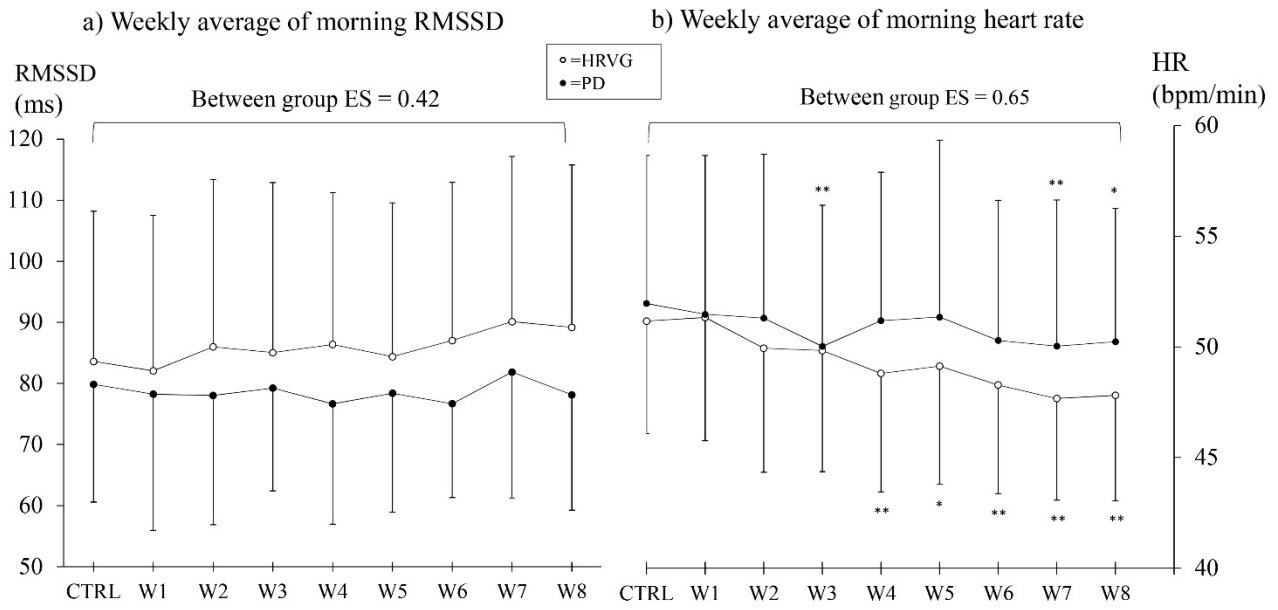
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2 **Figure 5.** Weekly average of morning RMSSD and heart rate. Statistical significance of within  
 3 group changes from the control period: \*  $p < 0.05$ , \*\*  $p < 0.01$

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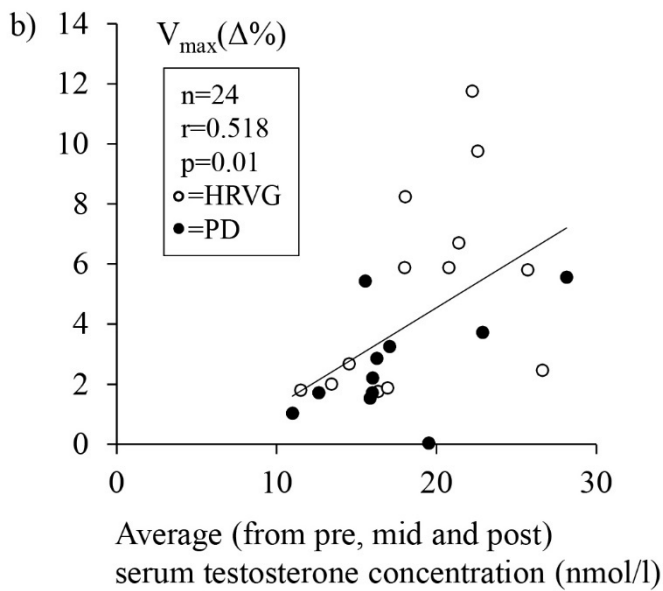
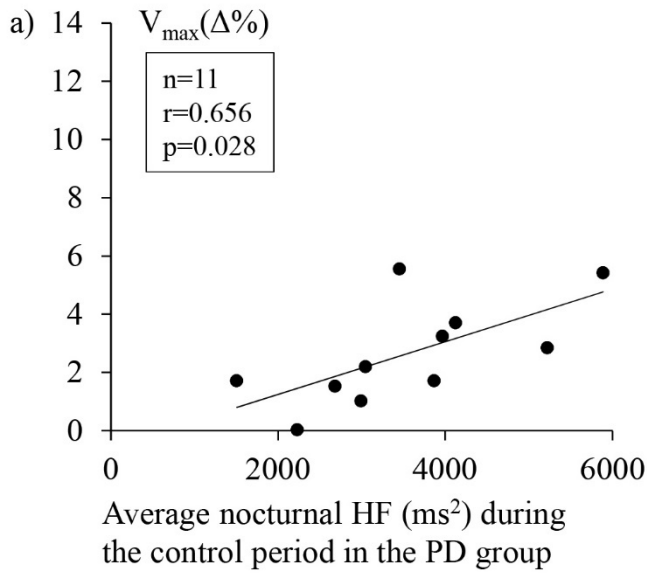
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2 **Figure 6.** a) Correlation between the absolute HF-values during the control period and relative  
 3 changes of  $V_{\max}$  in the PD group. b) Correlation between average serum testosterone concentration  
 4 at the pre, mid and post measurements and relative changes of  $V_{\max}$  in all subjects.

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1 **Table legends**

**Table 1. Training characteristic of both groups during the 8-week training period.**

	<b>HRVG (n=13)</b>	<b>PD (n=11)</b>
	<b>Weeks 1-8</b>	<b>Weeks 1-8</b>
<b>Training frequency (sessions/week)</b>	6.3 ± 1.4	6.1 ± 0.4
<b>Endurance training (h/week)</b>	5.1 ± 2.1	5.3 ± 1.8
<b>HRzone1 (%)</b>	82 ± 8	84 ± 7
<b>HRzone2 (%)</b>	15 ± 6	12 ± 5
<b>HRzone3 (%)</b>	3 ± 3	4 ± 3
<b>Other training (h/week)</b>	0.6 ± 0.2	0.7 ± 0.2
<b>Total training (h/week)</b>	5.7 ± 2.1	6.0 ± 1.9
Zone 1 < 82 %/HR <sub>max</sub> , zone 2 = 82-87 %/HR <sub>max</sub> and zone 3 > 87 %/HR <sub>max</sub> .		

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**Table 2. Incremental treadmill test results and between group effect sizes.**

	HRVG (n=13)		PD (n=11)		ES (pre-post)
	Pre	Post	Pre	Post	HRVG vs. PD
<b>VO<sub>2max</sub> (ml/kg/min)</b>	53.6 ± 4.2	56.7 ± 3.4**	54.2 ± 4.1	56.4 ± 4.7**	0.42 (small)
<b>VO<sub>2max</sub> (l/min)</b>	4.1 ± 0.3	4.3 ± 0.4*	4.0 ± 0.3	4.1 ± 0.3*	0.52 (moderate)
<b>V<sub>max</sub> (km/h)</b>	17.6 ± 1.3	18.5 ± 1.2***	18.0 ± 1.1	18.5 ± 1.2***	0.95 (large)
<b>LT1 (km/h)</b>	11.0 ± 1.5	11.8 ± 1.1*	11.6 ± 1.2	12.2 ± 1.2*	0.32 (small)
<b>LT2 (km/h)</b>	14.1 ± 1.0	15.0 ± 1.1***	14.7 ± 0.9	15.3 ± 1.2*	0.37 (small)
Statistical significances within group changes: * p<0.05, ** p<0.01, *** p<0.001					

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**Table 3. Running performance, serum hormone concentrations and heart rate variability (HRV) pre, mid and post. Effect size of between group differences was analysed from pre to post. Hormones were measured at the same day as the running test was performed. HRV was analysed as average of the control period (pre), week 4 (mid) and week 8 (post).**

	HRVG (n=13)			PD (n=11)			ES (pre-post)
	Pre	Mid	Post	Pre	Mid	Post	HRVG vs. PD
<b>Running</b>							
3000 m (min:s)	11:13 ± :50	10:52 ± :49***	10:38 ± :52*** <sup>ab</sup>	11:07 ± :47	10:42 ± :42**	10:32 ± :41** <sup>ab</sup>	0.00 (trivial)
MaxLa (mmol/l)	14.2 ± 3.0	14.3 ± 3.4	15.8 ± 2.9* <sup>b</sup>	12.4 ± 2.6	13.6 ± 3.0	13.9 ± 1.7	
MaxHR (bpm/min)	187 ± 9	186 ± 7	187 ± 7	193 ± 6	192 ± 7	192 ± 7	
<b>Hormones</b>							
Testosterone (nmol/l)	19.0 ± 5.3	17.7 ± 4.8	20.6 ± 4.8* <sup>b</sup>	18.3 ± 5.6	16.5 ± 4.3*	17.2 ± 4.3	0.59 (moderate)
Cortisol (nmol/l)	446 ± 70	464 ± 91	488 ± 109	458 ± 79	474 ± 50	482 ± 65	0.03 (trivial)
Testosterone/cortisol	0.43 ± 0.11	0.39 ± 0.13	0.45 ± 0.14	0.42 ± 0.12	0.35 ± 0.10	0.36 ± 0.09	0.24 (small)
<b>Heart rate variability</b>							
HR (bpm/min)	50.9 ± 5.6	48.9 ± 5.5	46.5 ± 5.0** <sup>a</sup>	52.2 ± 5.4	49.9 ± 5.8*	48.6 ± 5.5** <sup>a</sup>	0.20 (small)
RMSSD (ms)	76 ± 25	80 ± 22	89 ± 22* <sup>a</sup>	67 ± 12	72 ± 19	81 ± 29	0.10 (trivial)
LF (ms <sup>2</sup> )	4898 ± 1415	5438 ± 1532	6232 ± 2090* <sup>a</sup>	5165 ± 1904	5830 ± 2301	5768 ± 2204	0.65 (moderate)
HF (ms <sup>2</sup> )	4055 ± 2313	4324 ± 2177	4865 ± 2085	3542 ± 1210	3876 ± 1552	4163 ± 1923	0.36 (small)
TP (ms <sup>2</sup> )	8952 ± 3265	9762 ± 3208	11097 ± 3814* <sup>a</sup>	8707 ± 2727	9706 ± 3342	9931 ± 3353	0.43 (small)
Statistical significance within group changes: * p<0.05, **p<0.01. <sup>a</sup> =Pre-post, <sup>b</sup> =Mid-post							

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