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Heart rate variability and recovery following maximal exercise in endurance athletes and physically-active individuals

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Bentley *et al.* Athletic status and sympathovagal modulation**ABSTRACT**

The purpose of this study was to determine potential adverse cardiac effects of chronic endurance training by comparing sympathovagal modulation via heart rate variability (HRV) and heart rate recovery (HRR) in middle-aged endurance athletes (EA) and physically-active individuals (PA) following maximal exercise. 36 (53 ± 5 years) EA and 19 (56 ± 5 years) PA were recruited to complete a two-week exercise diary and graded exercise to exhaustion. Time domain and power spectral HRV analyses were completed on recorded R-R intervals. EA had a greater HRR slope following exercise (95%CI, 0.0134-0.0138 vs. 0.0101-0.0104 beats/second; $p < 0.001$). While EA had greater HRR 1-5 minutes post-exercise (all $p < 0.01$), PA and EA did not differ when expressed as a percentage of baseline HR (130 ± 19 vs. 139 ± 19 ; $p = 0.2$). Root mean square of successive differences in R-R intervals (rest and immediately post-exercise) were elevated in EA ($p < 0.05$). Low frequency (LF) and high frequency (HF) spectral components were non-significantly elevated post-exercise ($p = 0.045$ - 0.147) in EA while LF/HF was not different ($p = 0.529$ - 0.986). This data suggests greater HRR in EA may arise in part due to a lower resting HR. While non-significant elevations in HF and LF in EA produces a LF/HF similar to PA, absolute spectral component modulation differed. These observations require further exploration.

Novelty Bullets:

- Acute effects of exercise on HRV in EA compared to a relevant control group, PA, are unknown.
- EA had greater HRR, and non-significant elevations in LF and HF compared to PA, yet LF/HF was not different.
- Future work should explore the implications of this observation.

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Keywords: Low-frequency modulation, High-frequency modulation, Middle-aged endurance athlete, Time domain analysis, Power spectral analysis, Autonomic activity

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INTRODUCTION

Habitual endurance training produces distinct morphological adaptations to the heart, often termed the ‘Athlete’s Heart’ (Maron 1986). Additional adaptive changes are observed in sympathovagal balance, typically assessed as heart rate variability (HRV), a measure of the beat-to-beat variability in heart rate (HR), and HR recovery (HRR), a measure of the drop in HR following exercise after a fixed interval. Both HRV and HRR are accepted non-invasive means of exploring cardiac sympathovagal activation (Shetler et al. 2001; TaskForce 1996). In comparison to sedentary controls, athletes have faster HRR (Du et al. 2005), and a higher high frequency (HF) spectral component combined with lower HR both at rest (Du et al. 2005; Shin et al. 1995) and following acute exercise (Shin et al. 1995); with HRV measures mainly driven by parasympathetic modulation via the root mean square of successive differences in R-R intervals (RMSSD) (Uusitalo et al. 1996). Importantly, slower HRR and reductions in HRV are associated with autonomic dysfunction, increased mortality (Cole et al. 1999; Tsuji et al. 1996) and overreaching in athletes (Le Meur et al. 2013).

Some data suggest that vigorous prolonged exercise (Paffenbarger et al. 1986; Schnohr et al. 2015) performed with increasing frequency (Armstrong et al. 2015; Schnohr et al. 2015) well-beyond recommended levels, may increase the risk of adverse cardiovascular outcomes. A potential negative relationship is observed between exercise and autonomic function, as increased training loads reduce resting HRV within individuals (Manzi et al. 2009) while resting vagal-mediated HRV in highly trained individuals may be attenuated and appear to resemble that seen in sedentary rather than moderately-trained individuals (Buchheit et al. 2004). Despite the high participation rates of middle-aged athletes in endurance events (Running USA 2016), little is known about the effects of sustained exposure to exercise training on the sympathovagal

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contribution to HRV and HRR following an acute bout of exercise, a common daily perturbation in this cohort.

Therefore, the purpose of this study was to determine how chronic endurance training performed beyond recommended levels affects the pattern of HRV and HRR following maximal exercise in endurance trained athletes (EA) in comparison to a reference group of physically-active individuals (PA). We hypothesized that EA would have a faster rate of HRR following maximal exercise compared to PA. We further hypothesized that in comparison to PA, EA would have a greater RMSSD during the dynamic and stable HRR periods, and an elevated HF during stable HRR periods resulting in a reduced sympathovagal ratio.

MATERIALS AND METHODS

Participants

36 healthy middle-aged EA and 19 healthy middle-aged PA between 45 and 65 years of age participated in this study. Institutional research ethics boards approved this study (#30298) according to the terms of the Declaration of Helsinki and all participants initially provided written informed consent. Health status was confirmed by the completion for the Physical Activity Readiness Questionnaire for Everyone (PAR-Q+) (Jamnik et al. 2011). All EA were recruited from running, cycling or triathlon clubs with >10 years of regular competitive aerobic exercise participation in running, cycling and/or triathlon. Runners were completing weekly mileage in excess of 40 km with one or more annual marathons. Cyclists had weekly mileage exceeding 300 km with annual seasonal races of 100+ km. All PA were recruited from the community and performed up to 150 min/week of moderate to vigorous physical activity as per current guidelines (Tremblay et al. 2011), but did not engage in running competitions up to 10 km in distance with any regularity. We chose PA participants as a control group given the

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widely-accepted health benefits conferred by regular participation in recommended levels of physical activity, thereby reflecting an ecologically-valid reference group.

Study Design

Participants reported to the laboratory on two separate occasions. The first visit consisted of a standard anthropometric assessment of height and weight and the completion of a two-week exercise diary. The two-week exercise diary was completed in the laboratory in a standardized fashion utilizing exercise training notes and information that participants were instructed to record and collect throughout their training from the preceding two weeks. Body mass index (BMI) was computed as weight (kg) / height (m)². On the second visit, HRV and HRR following maximal exercise were assessed in addition to resting HRV and blood pressure (BP).

Maximal Exercise Test

Maximal exercise consisted of a graded treadmill exercise test to exhaustion and was typically completed within 8-12 minutes. Immediately following a 5 minute warm up at 5.6 km/hr, participants ran at a self-selected pace at 0% incline with grade increased by 2% every 2 minutes until 8 minutes, beyond which grade was increased by 1% every minute until volitional exhaustion (Weiner and Lourie 1969). Breath-by-breath pulmonary gas exchange was averaged every 20 seconds to obtain a peak rate of oxygen consumption ($\dot{V}O_{2peak}$; Moxus Metabolic Cart; AEI Technologies, Pittsburgh, Pennsylvania, USA).

Heart Rate and Blood Pressure

Resting seated BP (BpTRU model BPM-100, BpTRU Medical Devices, Coquitlam, BC, Canada) was measured in accordance with Canadian Hypertension guidelines (Harris et al. 2016) prior to assessing $\dot{V}O_{2peak}$. Participants were outfitted with a wireless HR monitor (Polar V800, Polar Electro Oy; Kempele, Finland) with R-R intervals continuously measured throughout rest,

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maximal exercise and 13 minutes (780 seconds) of recovery following exercise. Pre- and post-exercise measures of HR were performed in the seated position. Maximal BP during exercise was assessed with an automated auscultatory device on the right arm (Tango M2; SunTech Medical, Morrisville, North Carolina, USA).

Data Analysis

Prior to analysis, individual R-R intervals were processed and abnormal intervals were removed in accordance to the methods outlined by (Kemper et al. 2007) and were replaced using cubic spline interpolation.

Heart Rate Recovery

HR was recorded on a beat-by-beat basis with individual data linearly interpolated at 4Hz and averaged within EA and PA respectively. HR following exercise cessation was then plotted over 360 seconds (6 minutes) to capture HR plateau and subsequently averaged into 30-second time bins to assess HR stability for subsequent HRV analysis. HR was also averaged into 1-minute (60 second) time bins according to suggested guidelines (Adabag and Pierpont 2013) for HRR analysis and presented as HRR1 through to HRR5.

Heart Rate Variability

R-R intervals were imported into Kubios HRV analysis software (version 2.1, ©Kubios HRV, Kuopio, Finland). Time domain HRV analysis identifying RMSSD and the standard deviation of normal R-R intervals (SDNN) was completed. RMSSD was analyzed with ultra-short duration 30-second time periods (Baek et al. 2015) during 180 seconds of rest and during the dynamic portion of HRR following maximal exercise from 0-270 seconds. RMSSD represents vagally-mediated changes in HRV while SDNN in short-term recordings represents the parasympathetically-mediated respiratory sinus arrhythmia contribution to HRV (Shaffer and

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Ginsberg 2017). In accordance with the Task Force (TaskForce 1996), power spectral analysis was completed using a Fast Fourier Transformation in the presence of a stationary HR. Time domain and frequency domain assessments of HRV were completed from 480-780 seconds post-exercise. Welch's window was set at 256 seconds with an overlap of 50%. Low frequency (LF) spectral bands were defined as 0.04-0.15Hz while HF spectral bands were defined as 0.15-0.4Hz. R-R intervals were interpolated at 4Hz. LF bands represent a combination of sympathetic and parasympathetic modulation while HF bands represent parasympathetic or vagal modulation. LF/HF represents the balance of sympathovagal modulation (Shaffer and Ginsberg 2017). No artifact correction was applied within Kubios HRV and respiration rate at rest or following exercise was not controlled. RMSSD in 30-second time bins both at rest and following exercise from 0-270 seconds, and LF and HF from 480-780 seconds violated the assumption of normality. Data were Log transformed.

Exercise History

Exercise patterns were obtained from a two-week exercise diary with written descriptions of their current, routine exercise patterns. A Likert scale question on how the exercise diary related to 1, 5, and 10 years of exercise training was used to confirm a regular, long-standing exercise history. Weekly hours of vigorous endurance and resistive exercise were calculated and extrapolated to provide 10-year exercise hours, the minimum activity duration for participant inclusion.

Statistical Analysis

All statistics were calculated using a combination of SPSS 20 (IBM Corp), SigmaPlot 12.0 (Systat Software, Inc.) and GraphPad Prism 6 (GraphPad Software, Inc.). Normality of data was assessed visually with Q-Q plots and quantitatively with a Shapiro-Wilk test, with normally

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distributed variables assessed with independent samples t-tests. Mann-Whitney U tests were used to assess variables that violated the assumption of normality. HRR data was interpolated at 4Hz for each individual and then fit with a first order exponential decay within each group and compared with slope analysis. A mixed model repeated measures ANOVA was used to assess HRR in 1-minute intervals and to establish the onset of a stationary HR in 30-second time intervals. Log HF for EA had a Shapiro-Wilk test of exactly $p=0.05$. As such, frequency domain HRV data was analyzed using an independent samples t-test on Log transformed data, a Mann-Whitney U test on untransformed data, and a bootstrapped independent samples t-test on untransformed data. This analysis strengthened the confidence in the statistical outcome in light of the statistics surrounding normality. Parametric data are presented as mean $\pm$ SD. Non-parametric data are presented as median, interquartile range (Q1-Q3). Bootstrapped data are presented as 95% confidence intervals. Only significant F-statistics within the repeated measures ANOVA were further assessed using Bonferroni corrected *post hoc* tests. Assumptions of normality and homogeneity of variance in the repeated measures ANOVA were met. When the assumption of sphericity was violated a Greenhouse-Geisser correction was applied. Multiple linear regression analyses were performed with the application of a Bonferroni correction to the level of statistical significance. Statistical significance was set at $p<0.05$ for all analyses.

RESULTS

Participant Characteristics

Demographic assessments are presented in Table 1. EA had a lower resting HR ($p=0.009$) and BMI ($p=0.004$) compared to PA. As expected, EA presented with greater exercise training metrics (all $p<0.001$) and had a higher $\dot{V}O_{2peak}$ ($p<0.001$). PA had greater weekly hours of resistive exercise compared to EA ($p=0.047$). HR and BP at maximal exercise did not differ

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between EA and PA (all $p>0.1$).

Heart Rate Recovery

Maximal exercising HR was not different between EA and PA ($p=0.47$). Following maximal exercise, slope analysis revealed faster HRR in EA compared to PA (Figure 1 Panel A). EA had a greater slope (95% CI, 0.0134-0.0138 beats/second, $r^2=0.98$ vs. 0.0101-0.0104 beats/second, $r^2=0.99$) and a reduced tau (95% CI, 72.7-74.8 seconds vs. 96.4-98.6 seconds) (both $p<0.001$). HRR plateaued at 270 seconds (4.5 minutes) post-exercise and EA had a lower HR at 360 seconds (6 minutes, $p=0.009$) and 780 seconds (13 minutes, $p=0.003$). At 360 seconds, the percentage HR from baseline was not different in EA and PA (138 ± 19 vs. 131 ± 19 %, $p=0.2$), while percent recovery from peak was greater in EA (53 ± 5 vs. 47 ± 6 bpm, $p<0.001$) (Figure 1 Panel B). HRR was significantly greater in EA at all assessed time points (all $p<0.01$). Within EA and PA, HRR was greater than the previous HRR at all time points (all $p<0.001$) except HRR5 vs. HRR4 (both $p>0.1$) (Figure 1 Panel A). A positive linear relationship existed between resting HR and HR at the end of recovery ($r=0.715$, $p<0.001$; Figure 2 Panel A).

Heart Rate Variability

At rest (1.50 ± 0.20 vs. 1.36 ± 0.21 , $p=0.015$) and during the dynamic phase of HRR (immediately following the cessation of exercise until 270 seconds; 0.98 ± 0.28 vs. 0.81 ± 0.20 , $p=0.006$) there was a main effect of athletic status when comparing RMSSD across 30 second intervals with EA having greater RMSDD than PA. During the stationary phase of HRR from 480-780 seconds post-exercise, EA had greater RMSSD and SDNN in comparison to PA, while HR was lower (Table 2). Non-parametric assessments demonstrated no difference in LF ($p=0.15$), HF ($p=0.12$) and LF/HF ($p=0.7$). Parametric assessments on Log transformed data

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revealed a non-significant elevation in LF ($p=0.052$), and HF ($p=0.045$) in EA compared to PA and no difference in LF/HF ($p=0.7$). Finally, bootstrapped comparisons revealed a non-significant increase in LF ($p=0.059$) and HF ($p=0.059$) in EA compared to PA while LF/HF was not different ($p=0.53$) (Table 2). Correlational analysis revealed a positive linear relationship between resting RMSSD and post-exercise stable RMSSD ($r=0.439$, $p=0.001$; Figure 2 Panel B).

DISCUSSION

In this study we compared sympathovagal modulation via HRR and HRV in middle-aged EA and PA following maximal exercise. The key findings of the present study were: 1) Despite an elevated HRR in EA, HRR in relation to baseline was not different between EA and PA. 2) EA presented with increased RMSSD during rest and both the dynamic and stable phases of HRR. 3) EA had non-significant elevations in LF and HF power during the stable phase of HRR while the LF/HF ratio was not different. The results from this study align with our hypothesis that EA would have elevations in HF modulation acutely following exercise facilitating a faster HRR. An important novel finding was the elevated LF in EA, such that EA and PA had a similar degree of sympathovagal (LF/HF) balance, although at different absolute operating ranges.

Demographic Assessment

EA presented with a reduced resting HR and BMI compared to PA. Reductions in resting HR were expected as vagal activity has been shown to be elevated with training (Sandercock et al. 2005). A reduced BMI was not expected, but is congruent with the increased exercise levels in EA compared to PA. While research suggests that BMI is correlated with HRR1 (Barbosa Lins et al. 2015), we did not observe a relationship between BMI and HRR1 ($r=0.2$, $p=0.2$); however, this may be explained by the presence of a normal BMI range within our study population as opposed to a range of BMI from normal to obese (Barbosa Lins et al. 2015). PA

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were exercising approximately in accordance to Canadian physical activity guidelines (150 minutes of moderate to vigorous physical activity each week) (Tremblay et al. 2011), while EA were performing approximately three times this amount. As expected, EA's cardiorespiratory fitness (90th percentile) was greater than PA (60th percentile) (Pescatello 2014). We are confident in our classification of EA and PA and that these individuals reflect a representative sample of endurance trained athletes and physically-active individuals.

Heart Rate Recovery

In the present study we observed a unique HRR response in EA compared to PA. When interpreted in isolation, EA have a faster HRR in comparison to PA. Appreciating differences in fitness, this aligns with previous work comparing trained and untrained participants (Darr et al. 1988; Du et al. 2005) as well as across fitness levels (Cole et al. 1999; Trevizani et al. 2012). Interestingly, a study of master athletes with similar age and fitness levels to our population did not have faster HRR1 or HRR2 in comparison to sedentary controls but did have higher HRR at minutes 3-5 (Kwon et al. 2016). Some evidence suggests that HRR1 is related to training load (Buchheit and Gindre 2006) while others suggest it is linked to cardiorespiratory fitness (Lee and Mendoza 2012) and exercise performance (Lamberts et al. 2009). In any case, elevations in the independent variable are correlated with faster HRR. While we did not observe a relationship between HRR1 and physical activity ($r=-0.15$, $p=0.3$), we did observe a correlation between $\dot{V}O_{2peak}$ and HRR1, in which those with greater cardiorespiratory fitness have faster HRR ($r=-0.28$, $p=0.04$). Importantly, both EA and PA were still well above the normal cutoff representing an abnormal response at HRR1 (Cole et al. 1999).

Our HRR results become particularly interesting when scaled to resting HR. Within this context, EA and PA no longer differ in the relative degree of HRR. While EA present with a

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lower resting HR and a greater relative change from peak HR compared to PA, the relative change from baseline at the end of dynamic HRR is not different. Although not analyzed in this fashion, our results are congruent with a prior report examining younger female marathon runners compared to untrained controls (Du et al. 2005).

The hallmark post-exercise recovery response is characterized by an immediate parasympathetic (vagal) reactivation coupled with a slower withdrawal of sympathetic activity as the metabolic demands of exercise are mitigated (Imai et al. 1994). Indeed, EA demonstrated elevated RMSSD during the dynamic period of HRR in comparison to PA, suggesting that the chronic endurance training of EA promotes parasympathetic reactivation following maximal exercise. Despite differences in peak absolute exercise intensity between EA and PA, literature suggests a relative intensity dependence of HRR (Imai et al. 1994), thus allowing for an EA vs. PA comparison. In light of the lack of difference between EA and PA when HRR was scaled to resting baseline, it is possible that the degree of parasympathetic reactivation, and therefore HRR, is entirely proportional to resting HR. This would facilitate a similar degree of HRR over a fixed time frame despite similar peak heart rates. This is confirmed with correlational analysis between HR_{REST} and average RMSSD during dynamic HRR ($r=-0.61$, $p<0.001$) and HR_{REST} and HRR2-5 (r range 0.37-0.45, all $p<0.01$). Further HR_{REST} and HR at recovery are positively correlated ($r=0.715$, $p<0.001$; Figure 2 Panel A). While our work identified that 37% and ~17% of RMSSD and HRR respectively are explained by HR_{REST} , additional work is required to further identify contributing factors to these integrated relationships.

Heart Rate Variability – Time Domain

Time Domain HRV was assessed at rest and during both the dynamic and stable time period of HRR. At rest and during the dynamic component up to 270 seconds post-exercise, EA

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had greater RMSSD compared PA. Whilst it is recommended that time domain HRV analysis be completed on 5-minute windows (TaskForce 1996), recent work has identified the use of ultra-short duration 30-second time periods as a valid assessment tool for RMSSD (Baek et al. 2015), which provides greater temporal resolution within the context of acute recovery. The presence of an elevated RMSSD aligns with the faster HRR observed in EA and also corroborates the elevation in RMSSD and SDNN observed during stationary HRR. We observed an increased SDNN 480-780 seconds post-exercise, yet Du *et al.* (Du et al. 2005) did not observe a difference between endurance trained and control participants following maximal exercise. Previous work suggests that increasing exercise training load reduces resting SDNN and RMSSD to that of sedentary individuals (Buchheit et al. 2004). Our results suggest that following maximal exercise, middle-aged EA with a history of performing high training volumes have greater indices of parasympathetic modulation. An important corroboratory observation is individuals who present with greater RMSSD at rest also demonstrate greater RMSSD following maximal exercise.

Heart Rate Variability – Frequency Domain

Frequency domain analysis was assessed through a series of statistical approaches. LF and HF distributions violated the assumption of normality and values were Log transformed. However, upon transformation of HF in EA, HF had a Shapiro-Wilk normality assessment of exactly $p=0.05$. We were hesitant to apply strictly a parametric assessment and therefore used a more robust statistical approach by combining both parametric and non-parametric analyses as well as bootstrapping. Therefore, we are confident in reporting that there is a non-significant increase in LF and HF in EA compared to PA, while LF/HF is not different. Given the present level of variability, we were sufficiently powered to detect a minimum difference in LF of 81

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ms² and HF of 13.5 ms² (n=55, $\alpha=0.05$ $\beta=0.2$, Power=0.8). We were therefore slightly underpowered to statistically detect the observed difference between EA and PA in LF of 74±149 ms² and in HF of 12±25 ms², but the physiological difference and the resulting impact on LF/HF is readily apparent.

Our data reveals that elevated indices of parasympathetic modulation in EA, which are known to occur following endurance exercise training (Carter et al. 2003), is supported by our time domain HRV analysis and the non-significant elevation HF. Notwithstanding, we observed an unexpected non-significant increase in LF modulation. This aligns with a prior report of highly-trained individuals compared to moderately trained (Buchheit et al. 2004), yet no such increase was observed in a study of female marathon runners compared to sedentary individuals (Du et al. 2005). Increasing exercise training burden has been known to increase the LF component of HRV (Manzi et al. 2009) and resting muscle sympathetic nerve activity has been shown to be elevated in athletes (Ng et al. 1994), in addition to cardiac specific sympathetic activity (Neri Serneri et al. 2001). It is therefore suggestive that while LF may represent a contribution of sympathetic and parasympathetic modulation (Shaffer and Ginsberg 2017), it is likely that elevations in sympathetic modulation, not simply elevations in parasympathetic modulation, contributed to the non-significant increase presently observed in LF. In all the aforementioned studies, the assessments of HRV were completed only at rest and not following exercise. Our inclusion of post-exercise measures, while technically challenging, is novel and expands upon previous investigations. One can postulate that an elevated sympathetic modulation following exercise may facilitate an elevated inotropic effect that serves to maintain cardiac output and defend mean arterial BP in the face of bradycardia and peripheral vascular dilatation, which reduces vascular resistance during exercise. While EA presented with elevated

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indices of parasympathetic and sympathetic modulation, sympathovagal (LF/HF) balance between EA and PA was not different. To this end, the concurrent elevation in sympathetic modulation in EA may serve to maintain peripheral perfusion and potentially facilitate enhanced removal of metabolite accumulation. While this result suggests that chronic endurance training is not deleterious from a cardiac sympathovagal balance perspective, the effects of chronic elevations in autonomic activity are unknown. The practical implications of a shift in sympathovagal balance in healthy athletes have yet to be identified. Further investigation is warranted in order to clarify if these responses reflect a more complex adaptive response to exercise training than is currently understood, or if they reflect a 'tipping point' beyond which a maladaptive response to chronic, excessive exercise may occur.

Experimental Challenges and Limitations

Breathing rate was not controlled for when assessing HRV and may have influenced HRV. However, the very nature of controlling breathing rate, even at resting frequencies, also modulates HRV (Weippert et al. 2015). While the expected recovery ventilation may result in slight decreases in HF and increases LF respectively (Weippert et al. 2015), the relative effect would be equivalent between EA and PA. Some evidence suggests normalizing HRV measures to average HR in humans (Sacha 2013) or animal models with HR ranges of ~40 to 160 bpm (Billman 2013). Our two groups differed by ~8 bpm at rest and ~9 bpm during stable HRR. This normalization had no impact on the interpretation of our data. Some literature suggests that HRR after two minutes is modulated by exercise intensity and the presence of metabolites (e.g. plasma epinephrine, lactate, hydrogen ions and inorganic phosphate) (Perini et al. 1989). While all individuals provided a maximal effort, metabolites, both production during exercise and clearance following exercise, were not measured and may influence HRR. Our analysis did not

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examine biological sex effects within EA and PA groups. Females have greater parasympathetic and reduced sympathetic activity in both younger and older athletes (Gregoire et al. 1996), yet our preliminary analysis showed no sex-differences for either time or frequency domain assessments of HRV, aligning with previous work in an athletic, older cohort (Brown and Brown 2007). Future investigations are warranted to fully understand potential sex differences in HRV and clarify discrepancies within the literature. While detailing the exact modulation of the parasympathetic and sympathetic branches of the autonomic nervous system from HRV remains challenging, it is clear that are our EA and PA groups fundamentally differ in their respective LF and HF modulation. Future work combining non-invasive HRV assessments with invasive muscle sympathetic measures post exercise and or parasympathetic/sympathetic blockade are required clarify autonomic relationships and contributions to HRR. Finally, we completed frequency analysis for only one 5-minute time interval from 480-780 seconds post-exercise, precluding assessment of the temporal responses of HRV in recovery from maximal exercise. The present time selection was chosen to maximize the sample and statistical power of our analysis while conforming with the Task Force (TaskForce 1996). Nonetheless, our results offer novel insights into the autonomic contribution to HRR and HRV immediately post maximal exercise.

CONCLUSION

Our study demonstrates that EAs engaged in approximately 300% more than the recommended amount of weekly vigorous physical activity have elevated indices of parasympathetic modulation both at rest and acutely following maximal exercise, facilitating vagal reactivation after exercise that is highly related to resting HR. Additionally, a key novel finding is that EA also demonstrate non-significant elevations in LF modulation following

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exercise. This produces a LF/HF balance later in recovery that is not different from PA. These results suggest that EA do not experience alterations in sympathovagal balance stemming from chronic endurance training as assessed by HRV following maximal exercise, but do differ in absolute HRR due to a lowered resting HR. Future work is required to understand the effects of the elevated sympathetic modulation concomitant with increases in parasympathetic modulation in EA and whether or not it reflects a complex adaptive component of cardiovascular regulation, or a maladaptive response to sustained excessive exercise that may contribute to adverse outcomes in the long-term.

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STATEMENT OF CONFLICT OF INTEREST

The authors do not have any conflicts of interest to declare.

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Bentley *et al.* Athletic status and sympathovagal modulation**Table 1:** Demographic Information

Variable	EA (n=36)	PA (n=19)
Male/Female	22/14	8/11
Age (years)	53 $\pm$ 5	56 $\pm$ 5
Height (cm)	174 $\pm$ 11	169 $\pm$ 11
Weight (kg)	71 $\pm$ 14	73 $\pm$ 13
BMI (kg/m ²)	23.1 $\pm$ 2.6*	25.5 $\pm$ 3.3
HR _{REST} (bpm)	60 $\pm$ 11*	68 $\pm$ 11
HR _{MAX} (bpm)	171 $\pm$ 13	169 $\pm$ 12
SBP _{REST} (mmHg)	113 $\pm$ 12	113 $\pm$ 13
DBP _{REST} (mmHg)	73 $\pm$ 9	72 $\pm$ 9
SBP _{MAX} (mmHg)	205, 188-216	208, 192-216
DBP _{MAX} (mmHg)	79, 71-85	87, 73-101
$\dot{V}O_{2peak}$ (ml/kg/min)	48.4 $\pm$ 8.5*	36.1 $\pm$ 8.3
Weekly vigorous endurance exercise hours	7, 5-9*	1, 0-2
Weekly vigorous resistive exercise hours	0.3, 0-1.1*	1, 0.3-2
10-year endurance vigorous hours	3439, 2675-4812*	550, 0-1144
10-year combined vigorous hours	3982, 3259-4812*	1820, 834-2102

Parametric variables are presented as mean $\pm$ SD. Non-parametric variables are presented as median, interquartile range. BMI; body mass index, HR; resting heart rate, SBP; resting systolic blood pressure, DBP; resting diastolic blood pressure, $\dot{V}O_{2peak}$; peak rate of oxygen consumption.

* denotes statistically significant difference between EA and PA, $p < 0.05$.

Bentley *et al.* Athletic status and sympathovagal modulation**Table 2:** Time and Frequency Domain HRV Analysis During Stable HRR

Parameter	EA (n=36)	PA (n=19)	p-value
Time Domain			
HR (bpm)	80 ± 10	89 ± 12	0.007
RMSSD (ms)	11.0 ± 4.5	7.6 ± 3.4	0.005
SDNN (ms)	25.5 ± 9.0	20.2 ± 13.7	0.042
Frequency Domain			
LF (ms ²)	129, 57-242	72, 32-177	0.147
Log LF	2.1 ± 0.4	1.8 ± 0.5	0.052
Bootstrap LF (ms ²)	130 - 247	69 - 154	0.059
HF (ms ²)	24, 10-44	16, 3-36	0.119
Log HF	1.3 ± 0.5	1.0 ± 0.6	0.045
Bootstrap HF (ms ²)	23 - 41	12 - 27	0.059
LF/HF	5.0, 3.3-14.2	5.4, 3.7-8.1	0.986
Log LF/HF	0.77 ± 0.48	0.81 ± 0.51	0.773
Bootstrap LF/HF	6.9 - 13.8	5.2 - 36.0	0.529

Parametric variables are presented as mean ± SD. Non-parametric variables are presented as median, interquartile range. Bootstrapped variables are presented as 95% confidence intervals. HR; heart rate, RMSSD; root mean square of successive differences in R-R intervals, SDNN; standard deviation of normal R-R intervals, LF; low frequency, HF; high frequency.

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FIGURE CAPTIONS

Figure 1: Heart rate recovery. *Panel A:* Interpolated beat-by-beat heart responses for EA and PA with slope analysis and 95% confidence intervals. Error bars have been omitted except at baseline, peak and end recovery. *Panel B:* Percentage heart of baseline and percentage heart rate recovery from peak. HRR; heart rate recovery, 1,2,3,4,5; minute of recovery respectively. * denotes a statistically significant difference between EA and PA, $p < 0.05$. † denotes statistically significant difference between previous HRR within a group, $p < 0.05$.

Figure 2: Heart rate recovery and time domain heart rate variability. *Panel A:* Correlation between resting heart rate and heart rate during the stable phase of heart rate recovery following maximal exercise. *Panel B:* Correlation between average resting RMSSD and RMSSD during the stable phase of heart rate recovery following maximal exercise.

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