ORIGINAL RESEARCH ORİJİNAL ARAŞTIRMA

DOI: 10.5336/sportsci.2025-111397

Short-Term Autonomic Responses to Repeated Sprint Exercise Under Normobaric Hypoxia: A Randomized Controlled Trial

Normobarik Hipokside Tekrarlı Sprint Egzersizine Kısa Süreli Otonom Sinir Sistemi Yanıtları: Randomize Kontrollü Çalışma

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ABSTRACT Objective: Exposure to hypoxia during exercise intensifies physiological stress, potentially altering autonomic nervous system activity. In exercise programs that incorporate additional physiological stressors such as hypoxia, it is important to allow sufficient recovery time between training sessions to ensure that high-intensity efforts, such as sprints, can be performed at maximal capacity. This study examined the short-term autonomic effects of a single session of repeated sprint training in normobaric hypoxia (RSH), with a specific focus on heart rate variability (HRV) responses using time- and frequency-domain parameters to determine the sufficient recovery duration. **Material and Methods:** Eighteen moderately trained males (age: 19.35±1 y) were assigned in a randomized parallel-group design. Participants performed a cycling-based repeated sprint protocol under normoxic or hypoxic (FiO_2=14.5-14.7%) conditions. The protocol included 4 sets of 5×5-second sprints, separated by 25 seconds of rest and 5 minutes of active recovery between sets. HRV was measured during exercise and at rest across 4 time points: baseline, intervention day, 48h, and 72h post-intervention. **Results:** Hypoxia exposure significantly reduced standard deviation of normal-to-normal intervals during both exercise (p=0.017) and recovery (p=0.023), and was associated with a higher post-exercise heart rate (p=0.041). Additionally, resting HRV showed a significant time effect for root mean square of successive differences (RMSSD) (p=0.032), with both groups displaying increased parasympathetic activity at 72h post-exercise. At the end of the 72 period following the intervention, RMSSD values increased by 138% in the hypoxia group and by 76% in the control group. No statistically significant differences or interactions were observed in low frequency (LF), high frequency (HF), or the LF:HF ratio at any time point between the interventions. **Conclusion:** In summary, a single session of RSH elicited a greater increase in sympathetic activation and a more pronounced suppression of parasympathetic activity compared to the same training performed in normoxia, thereby delaying vagal recovery. These findings highlight the importance of carefully planning hypoxic training sessions with adequate recovery intervals to balance performance and recovery processes. Nonetheless, the results also suggest that a 72h rest period is sufficient for full autonomic recovery, as indicated by HRV measures.

Keywords: Acute hypoxia; normobaric; repeated sprint; heart rate variability; autonomic nervous system

ÖZET Amaç: Egzersiz sırasında hipoksi maruziyeti, fizyolojik stresin şiddetini artırarak otonom sinir sistemi aktivitesinde değişikliklere yol açabilmektedir. Hipoksi gibi ek fizyolojik stres kullanılarak yapılan egzersiz programlarında, sprint gibi yüklenmelerin maksimum eforlarla yerine getirilebilmesi için 2 egzersiz seansı arasında yeterli toparlanma süresi konulması önemlidir. Bu çalışmada, tek seans normobarik hipokside tekrarlı sprint antrenmanının [repeated sprint training in normobaric hypoxia (RSH)] kısa vadeli otonom etkileri incelemiş ve özellikle kalp hızı değişkenliği (KHD) üzerindeki yanıtları zaman ve frekans alanı parametreleriyle yeterli toparlanma süresini belirlemek için analiz edilmiştir. **Gereç ve Yöntemler:** Rastgele paralel grup tasarımında, yaş ortalaması 19,35±1 yıl olan 18 orta düzeyde antrenmanlı erkek çalışmaya dâhil edilmiştir. Katılımcılar, bisiklet ergometresinde normoksik veya hipoksik (FiO_2=%14,5-14,7) koşullar altında bisiklet bazlı tekrarlı sprint protokolünü uygulamışlardır. Her biri 5×5 sn'lik sprintler ve 4 setten oluşan, sprintler arasında 25 sn dinlenme ve setler arasında yaklaşık 50 W'da 5 dk'lık aktif toparlanma süresi içeren bir protokol uygulatılmıştır. HRV ölçümleri; bazal ölçüm, girişim günü, 48 saat ve 72 saat sonrası olmak üzere 4 zaman noktasında yapılmıştır. **Bulgular:** Hipoksi maruziyeti, egzersiz (p=0,017) ve toparlanma sırası (p=0,023) normal atımlar arası intervallerin standart sapması değerlerini anlamlı düzeyde düşürmüş; ayrıca egzersiz sonrası kalp atım hızı daha yüksek bulunmuştur (p=0,041). Dinlenme durumunda KHD analizinde ardışık normal atımlar arası intervallerin farklılıklarının kareleri toplamının ortalamasının karekökü [root mean square of successive differences (RMSSD)] için zaman etkisi anlamlı bulunmuştur (p=0,032). Girişim sonrası 72 saat sonunda RMSSD değerinde hipoksi grubunda %138 ve kontrol grubunda %76 artış gözlenmiştir. Düşük frekans [low frequency (LF)], yüksek frekans in [high frequency (HF)] ve LF:HF oranında tüm zaman noktalarında istatistiksel olarak anlamlı bir fark ya da etkileşim bulunmamıştır. **Sonuç:** Sonuç olarak, hipoksik koşullarda yapılan tek seanslık tekrarlı sprint antrenmanı, normokside yapılan aynı antrenmana nispeten sempatik aktivasyonu daha fazla artırmış ve parasempatik aktiviteyi daha fazla baskılayarak vagal toparlanmayı geciktirmiştir. Bu sonuçlar, performans ve toparlanma süreçlerinin dengelenebilmesi için hipoksik antrenmanın yeterli toparlanma süreleriyle dikkatli planlanması gerektiğine işaret etmekle birlikte 72 saatlik dinlenme süresinin KHD için tam toparlanma bakımından yeterli olduğunu göstermiştir.

Anahtar Kelimeler: Akut hipoksi; normobarik; tekrarlı sprint; kalp hızı değişkenliği; otonomik sinir sistemi

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Peer review under responsibility of Turkiye Klinikleri Journal of Sports Sciences.

Received: 19 Apr 2025 ***Received in revised form:*** 24 May 2025 ***Accepted:*** 13 Jun 2025 ***Available online:*** 09 Jul 2025

The heart rate variability (HRV), which enables the observation and evaluation of autonomic nervous system (ANS) activity, has gained widespread application in both clinical and athletic contexts.[1,2] It is utilized not only for assessing the general health and physiological condition of patients and athletes, but also as a practical tool for evaluating exercise's physiological responses to training, and identifying early indicators of overtraining.[3-5] In order to evaluate the responses to training loads, ANS activities can be observed by measuring HRV.[6] This non-invasive method provides prognostic information and is easy to apply and reproduce.[7-9] During physical exercise, increased activity of the sympathetic nervous system (SNS) and decreased activity of the parasympathetic nervous system (PNS) lead to cardio-acceleration, characterized by a reduced interval between the heartbeats.[7,10] Conversely, at rest, cardio-deceleration occurs, reflected in a lengthening of the interval between heartbeats. The vagus nerve acts as the primary mediator of parasympathetic stimulation to the heart and lungs. While aging is associated with a decline in vagal tone, regular exercise is positively correlated with its enhancement.[5] So, the SNS and PNS subsystems work in a coordinated manner to maintain sympathovagal balance.[11] While high frequency (HF) components detected by baroreceptors indicate parasympathetic dominance, whereas low-frequency components reflect sympathetic dominance.[11] ANS responses shift between sympathetic and parasympathetic activities based on exercise intensity and severity, resulting in varied outcomes in HRV. In particular, parasympathetic activity is suppressed immediately after high-intensity exercise but begins returning to baseline within 24 hours post-exercise.[12,13] In the context of exercise physiology, the low-frequency (LF) to HF ratio-a key marker of sympathovagal balance-progressively declines with longer exercise duration, with more pronounced decreases at higher intensities, reflecting a cumulative autonomic response.[14]

Several studies have examined the relationship between sprint training and HRV under hypoxic conditions. For example, one study applied repeated sprint training in hypoxia (RSH) to healthy young males at three simulated altitudes (1,500 m, 2,100 m, and 3,200 m) and found that the natural logarithm of the root mean square of successive differences (LnRMSSD) parameter was significantly higher during the recovery period following the Wingate test, but only at the 1,500 m altitude.[16] Similarly, Hamlin et al. reported a strong correlation between HRV indices and sprint performance after 6 RSH training sessions over a 3-week intervention period.[15] In their study, HRV parameters were lower during the intervention weeks in the hypoxia group compared to the normoxia group; however, physical performance improved progressively from week 1-3. These findings suggest that factors such as exercise type and intensity, altitude or hypoxia level, and the duration of the intervention period may influence HRV responses. Consequently, HRV measurement has been increasingly utilized in sports and exercise science as a non-invasive tool to monitor physiological stress, assess recovery processes, and guide training load adjustments.[17]

The aim of this study was to investigate the acute (during and immediately after RSH) and short-term (post-exercise recovery period) effects of RSH on HRV in moderately trained males. Both time-domain and frequency-domain HRV metrics were examined to enable a comprehensive evaluation of ANS regulation. The RMSSD, reflecting parasympathetic activity, and the standard deviation of normal-to-normal intervals (SDNN), indicating overall HRV, were considered as time-domain measures. In addition, frequency-domain parameters including LF:HF, and the LF:HF ratio were used to provide insights into rapid shifts in sympathetic and parasympathetic activity, which are particularly relevant during high-intensity intermittent exercise. By evaluating acute and short-term HRV responses, these complementary metrics were employed to generate evidence-based guidance for recommending optimal recovery durations after RSH, aiming to enhance the efficiency of this time-saving training method and offering practical implications for athletes, coaches, and practitioners.

MATERIAL AND METHODS

STUDY DESIGN

This study conducted in randomized parallel-group design [control group (CON) n=8 and hypoxia group

(HYP) n=8] and approved by Trabzon University Ethics Committee (date: October 22, 2024; 2024-10/2.1). A random numbering sequence corresponding to the number of participants was generated based on a pre-compiled list then participants were randomly assigned to intervention groups using a computer-based application. Throughout the study, the principles of the Declaration of Helsinki were upheld. Each participant was asked to read and sign a written informed consent form prior to participation. The design incorporated repeated measures over time to evaluate changes in HRV across different time points. Baseline HRV measurements were collected 1 week before the implementation of repeated sprint in normoxia or hypoxia exercise, and follow-up assessments were conducted at 48 and 72 hours post-intervention period.

Participants

Sixteen healthy, moderately trained male students (n=16; age: 19.35±1 y; height: 173.97±5.36 cm; weight: 72.65±8.39 kg) from the Faculty of Sport Sciences at Trabzon University participated in this study. The inclusion criteria determined as the follows; be non-smokers, be male, free from any chronic diseases, not using any medication, and without musculoskeletal injuries within the past 6 months. Additionally, participants must not have engaged in any training or resided at altitudes above 1,500 meters within the last 3 months and must have been training at least 3 days per week.

Participants were instructed to maintain their regular dietary habits and replicate the same meals prior to each testing and exercise session. Participants abstained from supplements to minimize confounding variables during the entire study period. Additionally, the intake of caffeinated or alcoholic beverages and foods was discouraged for at least 24 hours prior to each test and training session. To minimize confounding effects, participants were further advised to avoid engaging in any vigorous physical activity or exercise until the completion of the study. Due to changes in their schedules, 2 participants in the CON group withdrew from the study; therefore, data from a total of 14 participants (HYP: n=8; CON: n=6) were analyzed.

Repeated Sprint Test and Exercise

In the scope of baseline and intervention measurements; participants completed a cycling based repeated sprint protocol on a Monark 894E ergometer (Sweden), consisting of 4 sets of 5 sprints (5×5s) with 25 seconds of rest between sprints and 5 minutes of active recovery (~50 W) between sets. The pedal resistance determined as 7.5% of the participant's body mass. Before each session, participants performed a 5-minute warm-up at a low cadence (~50 W), which included a 5-second sprint at the end of each minute.[18]

The simulated altitude was induced via using the Everest Summit II Altitude Generator (Hypoxico Hypoxicator, New York, USA), which provided a respiratory gas mixture equivalent to an altitude of 3,000 meters (FiO_2=14.5-14.7). Prior to the initiation of the warm-up, a respiratory mask connected to the hypoxicator system was securely fitted to each participant's face. The interface was carefully adjusted and inspected for air leakage to ensure airtight integrity and the accurate delivery of the hypoxic gas mixture. Arterial oxygen saturation was monitored throughout the testing session using fingertip pulse oximetry (Hypoxico, New York, USA). All tests and measurements were conducted at sea level in the Trabzon Sports Performance Measurement and Ability Center.

Heart Rate Variability Measurement

HRV measurements were conducted via Polar RS800 (Kempele, Finland). The device was attached to participant chest in accordance with the user manual and HRV were initiated to record after participant rested 10 min in supine position. HRV records were continued up to 15 min after the repeated sprint test protocol. The collected data downloaded via Elite HRV were converted and analyzed as quantitative HRV data by using Kubios HRV Scientific Lite 4.1.2 software. Time- and frequency-domain analyses of HRV were performed. Artifact correction threshold of 5% was applied to ensure data quality. RR intervals were resampled at 4 Hz using equidistant interpolation. Signal detrending was conducted using the Smoothness Priors method, with a smoothing parameter set to 500 and a cutoff frequency of 0.035 Hz. All preprocessing procedures were conducted in accordance

with established standards for HRV analysis. The following parameters were evaluated; SDNN, RMSSD, normalized unit of LF power (Lfnu), normalized unit of HF power (Hfnu) and low frequency and high frequency power ratio (LF:HF). The mentioned HRV parameters were analyzed for the time points of resting period, during the repeated sprint exercise, immediately after the repeated sprint test and last 5 min of the 15 min of recovery period after the test. Participants visited the laboratory on four separate days: at baseline, on the day of the intervention, at 48 and 72 hours post-intervention. All measurements were carried out in the morning between 08:00-10:00.

STATISTICAL ANALYSIS

A priori power analysis was conducted using G*Power (version 3.1) for a repeated-measures analysis of variance (ANOVA) (within-between interaction), with α=0.05, power=0.90, and f=0.40 based on previous study reporting substantial HRV changes following RSH.[16] The analysis indicated a minimum sample size of 14 participants, which aligns with the final sample analyzed in this study. Statistical analysis of the data was performed using SPSS version 26. Data are presented as mean±standard deviation. The normality of data distribution was assessed using the Shapiro-Wilk test. HRV responses at baseline and during normobaric hypoxia and normoxia were compared using the independent samples Student's t-test for normally distributed variables, and the Mann-Whitney U test for non-normally distributed variables. To compare resting HRV data at baseline, day of intervention, and at 48 and 72 hours post-intervention, a one-way repeated measures analysis of variance (ANOVA) was used for normally distributed variables, while the Friedman test was applied for non-normally distributed variables. The assumption of sphericity was assessed using Mauchly's test of sphericity. In cases where the test indicated a violation ($p<0.05$), the Greenhouse-Geisser correction was applied to adjust the degrees of freedom for the repeated measures ANOVA. When a statistically significant difference was detected, Bonferroni "post hoc" tests were performed to identify pairwise differences. Partial eta squared (η^2) values were computed as a measure of effect size for the ANOVA analyses. Effect sizes were interpreted according to conventional thresholds: small ($\eta^2 \geq 0.01$), medium ($\eta^2 \geq 0.06$), and large ($\eta^2 \geq 0.14$). A significance level (α) of $p \leq 0.05$ was used to determine statistical significance for all analyses.

RESULTS

HRV COMPARISONS DURING AND AFTER REPEATED SPRINT EXERCISE

HRV indices were assessed during repeated sprint exercise and following a 15 minute recovery period, at 2 distinct time points across 2 separate testing days. Also, resting HRV values were evaluated 4 different time points. Group comparisons and associated statistical significances are reported below.

At rest, prior to repeated sprint exercise, there were no statistically significant differences between the HYP and CON groups across all HRV indices. Heart rate (HR) was similar in both conditions (HYP: 186.60±6.76 bpm vs. CON: 189.40±7.00 bpm; p=0.465). Similarly, no significant differences were observed for time-domain parameters (SDNN, RMSSD) and frequency-domain parameters (LFnu, HFnu, or LF:HF ratio) (all $p>0.05$).

At 15 minutes post of repeated sprint exercise, HRV parameters did not differ significantly between the groups. Although both SDNN (54.96±59.79 ms vs. 23.33±13.27 ms; p=0.950) and RMSSD (61.57±82.64 ms vs. 14.94±7.21 ms; p=0.852) were higher in the HYP group, these differences were not statistically significant ($p>0.05$). Similarly, no significant group differences were found in LFnu, HFnu, or the LF:HF ratio (all $p>0.05$).

During the RSH and repeated sprint number (RSN) interventions, HR responses remained statistically similar (p=0.223). However, a significant reduction in SDNN was observed in the HYP group compared to CON (9.8±4.06 ms vs. 17.75±6.63 ms; p=0.017), indicating a potential suppression of HRV under hypoxic condition. RMSSD was also lower in HYP (9.17±4.35 ms) versus CON (15.38±10.65 ms), though not statistically significant (p=0.491). Other HRV indices (LFnu, HFnu, LF:HF) did not differ significantly between groups (all $p>0.05$).

Following the RSH/RSN session end of the 15 min recovery period, significant group differences emerged. Heart rate remained elevated in the HYP group (99.140±9.01 bpm) compared to CON (85.27±13.68 bpm; p=0.041). Additionally, SDNN was significantly lower in HYP (13.31±4.50 ms) than in CON (27.70±14.90 ms; p=0.023), suggesting sustained autonomic imbalance post-exercise under hypoxic condition. Although RMSSD tended to be lower in HYP (6.95±4.38 ms) versus CON (16.08±8.89 ms), this difference was not statistically significant (p=0.081). There were no significant differences for LFnu, HFnu, or LF:HF ratio between conditions (all p>0.05).

The analyses for HRV parameters obtained during the baseline and intervention days are presented in Table 1.

RESTING HRV COMPARISONS

Resting HRV indices were compared across four different time points between the groups. The outcomes of the repeated measures ANOVA are reported for main effects of time, condition, and interaction.

Resting HR showed no significant main effects for time (p=0.250, η^2p=0.110), condition (p=0.368, η^2p=0.068), or time×condition interaction (p=0.786, η^2p=0.018). Although the main effect of time for SDNN approached statistical significance (p=0.069, η^2p=0.197), neither the main effect of condition (p=0.472) nor the time×condition interaction (p=0.605) reached significance. SDNN values demonstrated temporal variability, with a pronounced increase at 72 hours post-intervention in both groups, indicative of a delayed restoration of overall autonomic modulation.

A significant main effect of time was observed for RMSSD (p=0.032, η^2p=0.248), whereas neither the main effect of condition (p=0.472) nor the time×condition interaction (p=0.637) reached significance. “post hoc” Bonferroni comparisons indicated a statistically significant increase in RMSSD from post-48 to post-72 in both groups (p=0.02). Specifically, RMSSD increased from 40.55±22.07 ms to 96.48±71.88 ms in the HYP group, representing a 138% increase, and from 41.19±23.57 ms to 72.39±30.87 ms in the CON group, corresponding to a 76% increase. No significant differences were detected in LFnu or HFnu power across time (LFnu: p=0.131; HFnu: p=0.131), condition (both p>0.7), or interaction terms (p>0.59). The LF:HF ratio did not show any significant changes over time (p=0.231, η^2p=0.111), between conditions (p=0.855), or in interaction effects (p=0.719).

The analyses for HRV parameters assessed at rest across the 4 time points are summarized in Table 2.

TABLE 1: Heart rate variability during and 15 minutes after the repeated sprint exercise for baseline and normobaric hypoxia/normoxia conditions

		HR	SDNN	RMSSD	LFnu	HFnu	LF:HF
Baseline RSA	HYP	186.60±6.76	109.38±126.79	143.41±160.73	50.63±13.38	49.19±13.32	1.24±0.97
	CON	189.40±7.00	38.17±25.39	50.43± 36.19	45.04±9.91	54.78±9.89	0.89±0.47
	p value	0.465	0.282	0.282	0.491	0.491	0.491
Baseline Post RSA	HYP	97.08±15.29	54.96±59.79	61.57±82.64	80.43±22.06	19.46±21.89	12.42±11.80
	CON	91.53±13.78	23.33±13.27	14.94±7.21	85.33±15.45	14.63±15.41	14.97±14.41
	p value	0.491	0.950	0.852	0.755	0.755	0.722
During RSH/RSN	HYP	188.99±10.12	9.8±4.06	9.17±4.35	65.13±17.25	34.74±17.20	3.33±3.73
	CON	182.88±6.53	17.75±6.63	15.38±10.65	62.71±20.99	37.16±20.93	3.33±4.03
	p value	0.223	**0.017***	0.491	0.817	0.816	0.852
Post RSH/RSN	HYP	99.14±9.01	13.31±4.50	6.95±4.38	88.55±4.30	11.41±4.27	8.85±3.60
	CON	85.27±13.68	27.70±14.90	16.08±8.89	88.83±7.88	11.14±7.87	11.34±6.40
	p value	0.041*	**0.023***	0.081	0.933	0.935	0.573

*p<0.05. HR: Heart rate; SDNN: Standard deviation of normal-to-normal intervals; RMSSD: Root mean square of successive differences; LFnu: Normalized low frequency power; HFnu: Normalized high frequency power; LF:HF: LF:HF ratio; HYP: Hypoxia group; CON: Control group; RSA: Repeated sprint ability; RSH: Repeated sprint training in hypoxia; RSN: Repeated sprint number

TABLE 2: Resting heart rate variability comparisons for four different time points

		HRV time-points				ANOVA p value (η^2p)		
		Baseline day	Intervention day	Post-48	Post-72	Time	Condition	Interaction
HR	HYP	66.748±9.98	67.50±8.33	68.86±8.89	61.27±9.77	0.250 (0.110)	0.368 (0.068)	0.786 (0.018)
	CON	63.05±8.22	64.03±9.18	64.25±8.74	60.95±4.83			
SDNN	HYP	78.39±38.82	54.64±18.05	50.15±14.26	86.32±42.48	0.069 (0.197)	0.472 (0.044)	0.605 (0.042)
	CON	64.43±40.96	60.34±11.40	49.33±17.50	69.25±23.08			
RMSSD	HYP	78.17±60.94	44.98±24.86	40.55±22.07	96.48±71.88	**0.032** (0.248)	0.472 (0.044)	0.637 (0.037)
	CON	55.86±46.51	45.65±10.11	41.19±23.57	72.39±30.87			
LFnu	HYP	64.97±12.41	75.26±9.42	67.53±20.40	68.38±13.33	0.131 (0.143)	0.737 (0.010)	0.590 (0.051)
	CON	66.57±12.04	74.77±4.70	76.09±10.09	65.68±17.69			
HFnu	HYP	34.97±12.38	24.71±9.41	32.43±20.39	31.55±13.31	0.131 (0.143)	0.734 (0.010)	0.590 (0.051)
	CON	33.37±11.97	25.15±4.70	23.86±10.07	34.24±17.61			
LF:HF	HYP	2.32±1.66	3.62±2.01	3.09±1.99	2.73±1.62	0.231 (0.111)	0.855 (0.003)	0.719 (0.036)
	CON	2.32±1.12	3.1±0.86	4.09±2.74	2.64±1.85			

*p<0.05; η^2p: partial eta-squared. HRV: heart rate variability; ANOVA: Analysis of variance; HR: Heart rate; HYP: Hypoxia group; CON: Control group; SDNN: Standard deviation of normal-to-normal intervals; RMSSD: Root mean square of successive differences; LFnu: Normalized low frequency power; HFnu: Normalized high frequency power; LF:HF: LF:HF ratio

DISCUSSION

This study demonstrated that a single RSH session suppresses sympathovagal balance acutely (during and immediately after exercise), and also induces a significant short-term autonomic disturbance, followed by an enhanced vagal rebound at 72 hours. The findings revealed that while hypoxic condition did not significantly alter resting HRV across a 4 day observation period (except of RMSSD), RSH elicited a pronounced reduction in HRV-particularly in SDNN-during and shortly after the intervention. These outcomes suggest that hypoxic stress during high-intensity interval exercise imposes a transient autonomic disturbance, likely characterized by reduced vagal activity and delayed parasympathetic reactivation. Notably, both the HYP and CON groups demonstrated a marked increase in RMSSD between 48-72 hours post-exercise. Specifically, RMSSD increased from 40.55±22.07 ms to 96.48±71.88 ms in the HYP group, representing a 138% increase, and from 41.19±23.57 ms to 72.39±30.87 ms in the CON group, corresponding to a 76% increase. These findings suggest a substantial enhancement in parasympathetic modulation during the later recovery phase in both groups; however, the increase was more pronounced in the HYP group, indicating that high-intensity exercise performed under hypoxic condition may have a greater capacity to facilitate vagal reactivation. The results showed that a 72 hour recovery period after a RSH session provides adequate time for autonomic restoration, indicating that this duration may be appropriate for safely repeating high-intensity exercise under normobaric hypoxia condition. These findings highlight the value of HRV in tracking autonomic recovery, especially after hypoxia-based training that imposes greater physiological stress than normoxic conditions.

Consistent with Hamlin et al. who reported an 8.5% decrease in RMSSD and an 11.5% decrease in SDNN following a 3 week RSH protocol, our study also observed acute and short-term reductions in HRV during and after a single RSH session.[15] SDNN was significantly lower during exercise in the HYP group (9.8±4.06 ms) compared to the CON group (17.75±6.63 ms; p=0.017), while RMSSD was also reduced (9.17±4.35 ms vs. 15.38±10.65 ms), though this difference was not statistically significant. A previous study have reported reduced HRV in hypoxia interventions compared to normoxia despite unchanged performance, likely due to additional physiological stress.[19] Similarly, Hamlin et al. emphasized that RSH training should be applied cautiously to prevent overtraining, highlighting the importance of allowing sufficient recovery time between sessions.[15] Our findings further support this caution, as short-

term reductions in HRV observed after a single RSH session suggest that hypoxic training imposes considerable autonomic stress, even in the absence of immediate performance decrements.[19]

The present findings align in part with those of Gutknecht et al. who examined HRV and heart rate recovery (HRR) following Wingate tests performed at simulated altitudes of 1,500 m, 2,100 m, and 3,200 m.[16] Their results showed modest changes in HRV indices, such as LnRMSSD (e.g., 3,200 m: pre = 1.98±0.90, post = 1.89±0.58), and no significant group × time interactions for HRR or HRV, with small effect sizes (η^2p=0.01-0.06).

Additionally, while our data showed that RSH induced a pronounced acute and short-term autonomic disturbance, it also demonstrated that a 72 hour rest interval may be sufficient for recovery. This is in contrast to the study on elite badminton players, where no significant changes in HRV or creatine kinase were observed up to 48 hours post-session, regardless of whether training was conducted under normoxia, systemic hypoxia (FiO_2=14%), or with blood flow restriction (RS-BFR). Although the badminton players experienced greater perceived fatigue and reduced performance-particularly in RS-BFR and RSH conditions-the absence of significant autonomic or muscle damage markers suggests that the physiological strain may be more localized or sport-specific, and less systemic than in cycling-based repeated sprint protocols.[20]

The pattern of autonomic recovery observed in our study is further supported by the work of James et al. who investigated time- and frequency-domain HRV responses following moderate and severe intensity exercise in trained runners.[12] Their findings showed a significant reduction in HRV indices-particularly RMSSD and SDNN 1 hour after severe intensity exercise, followed by a gradual return to baseline between 24 and 72 hours post-exercise. Specifically, RMSSD declined from 72 ms to 33 ms at 1 hour post-exercise, before recovering to 69 ms by 72 hours. A similar trend was observed in SDNN, which dropped from 72 ms to 44 ms and returned to 71 ms over the same period. These delayed recovery dynamics align closely with our observations, where RMSSD increased by 138% between 48 and 72 hours following RSH. Together, these findings underscore that high-intensity or hypoxic exercise may induce extended autonomic disruption, requiring at least 48 to 72 hours for full parasympathetic restoration. This highlights the importance of recovery monitoring in hypoxic training protocols, particularly for tailoring session frequency and avoiding cumulative autonomic fatigue in athletes.

In the present study, involving repeated sprint cycling under normobaric hypoxia, HRV indicated short-term autonomic impairment with full recovery by 72 hours. Similarly, Buchheit et al. found that RMSSD required 72-96 hours to return to baseline after a competitive soccer match, reflecting significant autonomic stress from high-intensity intermittent exercise, consistent with our findings.[21] These parallel recovery timelines reinforce RMSSD as a reliable marker of autonomic restoration and highlight the need for sufficient recovery periods, particularly when training involves added stressors like hypoxia.[9,16,21] According to Oliveira et al. hypoxia can markedly disrupt autonomic balance through vagal suppression, sympathetic dominance, or both, with effects amplified in unacclimatized individuals and influenced by factors such as altitude severity and barometric pressure changes, illustrating the complex interaction between environmental stress and cardiovascular regulation.[22]

Buchheit et al. emphasized that maintaining consistent HRV monitoring alongside exercise variables-such as training volume, intensity, and exercise modality-can be a valuable strategy for determining appropriate load and recovery intervals.[9] In this context, integrating HRV assessments into regular training monitoring can help optimize the balance between stress and recovery, especially when applying hypoxic protocols with high intensity exercises. Supporting this, our findings demonstrated a significant short-term suppression of HRV following RSH, with full parasympathetic restoration occurring within 72 hours-highlighting HRV's sensitivity to both exercise intensity and recovery status.

Cámara et al. demonstrated that acute hypoxia without physical loading did not significantly alter time- or frequency-domain HRV parameters, suggesting that short-duration hypoxia alone may not

disrupt autonomic balance.[23] This reinforces the importance of exercise type, intensity, and volume in modulating autonomic responses under hypoxic conditions. Consistent with James et al. our study confirms that high-intensity intermittent exercise under hypoxia elicits greater autonomic disturbance than moderate-intensity efforts or hypoxia alone.[12] These findings, supported by previous research, highlight HRV as a sensitive marker for monitoring autonomic stress and recovery during hypoxic training.[12,15,21] The combination of intense exercise and hypoxia amplifies autonomic demands, as noted by Oliveira et al. and Cámara et al. with disruption influenced by altitude, exposure time, and individual acclimatization.[22,23] Therefore, HRV data should be interpreted alongside training load characteristics (e.g., session frequency, intensity, volume, recovery intervals) and environmental factors such as FiO_2 levels and altitude. Practitioners implementing RSH should allow at least 72 hours for full autonomic recovery, particularly when reductions in parasympathetic activity (e.g. decreased RMSSD) are observed. Tailoring training frequency and load based on individualized HRV trends may help mitigate fatigue, reduce overtraining risk, and optimize training efficiency. Given the variability in hypoxia responses, future research should focus on developing personalized RSH protocols guided by HRV and autonomic markers.[24]

According to findings of this study, which examined the acute and short-term changes in HRV induced by a single session of RSH training, demonstrated that a 72 hour rest period is sufficient for the recovery of observed HRV parameters in moderately trained males.

CONCLUSION

In conclusion, this study demonstrated that repeated sprint exercise under normobaric hypoxia induces a marked acute and short-term alterations of autonomic function, as evidenced by significant reductions in HRV parameters-particularly SDNN and RMSSD-during and immediately after exercise. However, the pronounced rebound in RMSSD at 72 hours post-intervention suggests that a 72 hour recovery period is sufficient to restore autonomic balance following RSH in moderately trained males. Nevertheless, future research is warranted to explore the cumulative autonomic stress imposed by multiple RSH sessions, particularly with respect to HRV observations during recovery.

Source of Finance

During this study, no financial or spiritual support was received neither from any pharmaceutical company that has a direct connection with the research subject, nor from a company that provides or produces medical instruments and materials which may negatively affect the evaluation process of this study.

Conflict of Interest

No conflicts of interest between the authors and / or family members of the scientific and medical committee members or members of the potential conflicts of interest, counseling, expertise, working conditions, share holding and similar situations in any firm.

Authorship Contributions

This study is entirely author's own work and no other author contribution

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