Central European Journal of Sport Sciences and Medicine | Vol. 53, No. 1/2026: 87–98
ISSN (print): 2300-9705 | ISSN (online): 2353-2807 | DOI: 10.18276/cej.2026.1-08

EFFECT OF AEROBIC FITNESS ON CARDIO-VAGAL ADAPTATION

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Abstract Introduction: Maximal oxygen consumption (VO_2 max) and heart rate variability (HRV) are important physiological indicators of both cardiovascular fitness and autonomic nervous system regulation in exercise science. This study aimed to examine the effect of aerobic fitness on cardio-vagal adaptation in young adults. Methods: Thirty-five physically active university students (23.89±1.64 years), with a minimum of four years of regular exercise participation were included in this study. VO_2max was assessed using a graded treadmill test and HRV indices were obtained from ECG recordings using LabScribe and Kubios software. Pearson's correlation with Bonferroni-adjusted was performed for statistical analysis. Results: No significant correlations were observed between VO_2 max and time-domain HRV indices SDNN(r=0.177, p=0.309); pNN50(r=0.198, p=0.254) and NN50(r=−0.340, p=0.846), while RMSSD(r=0.340, p=0.045) showed weak to moderate positive association. In contrast, significant correlations were found with frequency-domain indices, including HF power (r=0.450, p=0.006) and LF power (r=−0.450, p=0.006) and LF/HF ratio (r=−0.449, p=0.006). Conclusion: These findings indicate that regular aerobic exercises associated with improved parasympathetic modulation and enhanced cardio-autonomic balance in young adults.

Key words: cardio autonomic function, graded exercise protocol, heart rate variability, parasympathetic tone, VO2max

Introduction

According to the World Obesity Atlas 2024 published by the World Obesity Federation, it is projected that by 2035, over 1.77 billion individuals will be classified as overweight, while 1.53 billion will face obesity. This will account for 54% of the global adult population (Janić et al., 2025). Insufficient physical activity is recognized as the primary contributor to this obesity and is also a significant factor in mortality rates. In light of the recent increasingly prevalent technology-driven lifestyle, the rates of obesity continue to rise each year (Mittal et al., 2022).In this respect regular physical activity (PA) can be a non-pharmacological mediator for reduced obesity and improved cardiorespiratory fitness (CRF). The enhancement of CRF or aerobic fitness (VO2Max) for physical well-being through PA has been noted and documented in physically active individuals with normal weight (Chen et al., 2019; Alansare et al., 2021; Masroor et al., 2018).

An individual's physical as well as mental well-being is influenced by the proper release of hormones and a balanced autonomic nervous system. Any existing cardiometabolic issue disrupts the typical functioning of cardiac autonomic functioning (CAF) in the body, which can have health consequences (Oliveira et al., 2020; Silva et al., 2017; Rossi et al., 2015). Evaluating CAF through heart rate variability (HRV) assessment is considered a straightforward, non-invasive, and dependable approach (Ziemssen & Siepmann, 2019; Catai et al., 2020), and aerobic fitness is a crucial factor in sustaining cardiac autonomic health. While VO2 max (the maximum amount of oxygen consumed) is considered the premier assessment tool for CRF (Zoolaktaf, 2007), heart rate variability (HRV) measures the variation in time between successive heartbeats, which arises from an integrated regulatory system responding to various psychological and environmental stresses (Shaffer & Ginsberg, 2017). Engaging in regular aerobic training or exercise results in both structural and functional adaptations in the cardiovascular system, which may influence HRV measurements. These alterations are particularly beneficial in decreasing the likelihood of cardiovascular diseases and premature mortality, thereby positively impacting cardiovascular well-being and the operational state of the autonomic nervous system (ANS) (Sandercock et al., 2005; Hottenrott et al., 2006; Routledge et al., 2010).

Research studies have indicated that aerobic exercise programs can lead to an increase in heart rate variability (HRV) among healthy adults (Carter et al., 2003; Iwasaki et al., 2003; Tulppo et al., 2003). In line with this, various exercise interventions have been shown to improve HRV even in sedentary middle-aged individuals (Navarro-Lomas et al., 2022). A correlational study involving healthy young adults who did not engage in any rigorous physical activity demonstrated that VO2 max is linked to heart rate variability metrics (Saini et al., 2022)

While existing studies suggest that exercise boosts both VO_2 max and HRV, the relationship between these two elements in regularly active, non-clinical groups remains unclear. Therefore, additional investigation into how these factors interact is essential. Hence, the objective of the current study was to explore the relationship between VO2max and HRV indices in healthy young adults using a cross-sectional research approach.

Methods

Participants

The study was executed at Exercise and Sports Physiology Laboratory, Department of Physical Education, Jadavpur University, Kolkata, India. G*POWER software (Version3.1.9.7) was used for calculation of sample with effect size=0.50, α=0.05 and the power =0.85 (Faul et al., 2009; Kang, 2021). According to the G*POWER calculation the required sample size (n) was at least 32. Thus, a total 35 adult youth subjects aged 23.89±1.64 were recruited in the present study. Prior to commencing the study, clearance from institutional ethical committee was taken (Ref No: IEC/37/C/25) and for the recruitment of the participants, researchers organized an orientation program to give detailed explanation about the study. All interested candidates were requested to complete the personal consent form, and ultimately participants were selected via purposive sampling to fulfill the study's objectives. The study was also approved by the Clinical Trial Registry of India (CTRI) with registration number CTRI/2025/05/086537.

Inclusion and Exclusion criteria

Male participants aged between 23.89±1.64 years with minimum four years' regular involvement in physical activity or exercise at least five days in a week for two hours each day were assigned as subjects in this study. All the participants were selected from Jadavpur University, stayed in the hostel thus followed same biological clock and diet pattern. Individuals with any chronic health conditions, injuries, or medical disorders were not considered in the present study.

Administration of Tests

Researchers assessed aerobic fitness using the Treadmill Test (TMT) based on the Bruce Protocol, employing a complete iworx system that included the IX-TA-220 acquisition unit, iWire-GA1 CO_2/O_2 gas analyzer with filter, a 3-liter calibration syringe, flow-head tubing with an A-FH-1000 flow head, an A-GAK-201 reusable mask, a 5-liter mixing chamber (A- GAA-MC), an A-CAL-200 calibration kit, and the PHRM-220 heart-rate monitor—all manufactured by iworx Systems Inc. (Dover, NH, USA). The Bruce Protocol, widely used to estimate cardiorespiratory fitness, consists of seven three-minute stages, each increasing in treadmill speed and incline, and is designed to determine maximal oxygen uptake (VO_2 max). For the assessment of VO2 max participant's preparation involved several phases. In first phase (resting/supine), the skin was cleaned and mildly abraded to ensure good electrode contact, following this preparation, ten ECG electrodes were positioned on the participant using the standard Mason–Likar placement illustrated in Table 1. The acquisition system was then connected, and a resting ECG was recorded along with baseline measurements including heart rate and blood pressure, typically taken while the participant was lying supine. In second phase consisted of a brief standing baseline, allowing comparison between supine and upright physiological values before exercise. In third phase, participants performed controlled hyperventilation while standing to differentiate respiratory-related ECG variations from possible cardiac abnormalities. After all initial data were collected, participants were briefed about the test procedure, safety instructions were reviewed, and the respiratory mask was fitted securely and checked for comfort and proper seal.

Once all preparations were completed, the treadmill test began following the automated Bruce Protocol. Participants started walking, progressing through the increasing stages of speed and incline every three minutes, the complete detail of which is given Table 2 below. When the participant's heart rate reached 85% of their maximum, the test was stopped, and LabScribe software (version 4.0) was used to generated the individual VO_2 max values and final assessment report.

Table 1. Placement of Leads for the Assessment of VO2 max.

Lead	Placement
RA (Right Arm)	Below right clavicle, on the outer edge of the chest
LA (Left Arm)	Below left clavicle, on the outer edge of the chest
RL (Right Leg, Ground)	On the lower right abdomen or right iliac crest
LL (Left Leg)	On the lower left abdomen or left iliac crest
V1	Fourth intercostal space at the right sternal border
V2	Fourth intercostal space at the left sternal border
V3	Midway between V2 and V4
V4	Fifth intercostal space at the midclavicular line
V5	At the same horizontal level as V4, at the anterior axillary line
V6	At the same horizontal level as V4 and V5, at the midaxillary line

Table 2. Stages, Speed and Inclines of Bruce Protocol.

Stage	Treadmill speed	Treadmill incline
1	1.7 mph	10%
2	2.5 mph	12%
3	3.4 mph	14%
4	4.2mph	16%
5	5.0 mph	18%
6	5.5 mph	20%
7	6.0 mph	22%

For the assessment of HRV from electrocardiogram (ECG) the researchers used iwire-B3G instrument through LabScribe software (version 4.0). Before the assessment, the participants relaxed for half an hour in a quiet room with a constant temperature. Then they were made to sit comfortably on an armchair keeping both legs on the wooden cube, three disposable ECG electrodes were fixed on the underside of the right and left wrist subsequently and on the right ankle as well. All electronic and metal items were cautiously taken away from the participants, and ECG signals were recorded for 25 minutes. From this, artifact-free 5-minute segments were selected for R–R interval extraction. During R–R interval processing, researchers used ECG data filtered within the 500–1000 Hz range (Malik et al., 1996). Finally, the R–R interval data were imported into Kubios HRV Standard software (version 3.5.0), which generated the time- and frequency-domain HRV analysis. Full details of HRV variables were provided in Table 3 below whereas Figure 1 illustrates the trial profile and outlines the primary outcome.

Table 3. Details of Time Domain and Frequency Domain Variable of HRV.

	Variables	Description	Units	Dominance
TIME DOMAIN	RMSSD	The Root Mean Square of Successive Differences between normal heartbeats	milli seconds (ms)	Parasympathetic dominance
	SDNN	Standard Deviation of the N-N intervals	milli seconds (ms)	Parasympathetic dominance
	NN50	Adjacent NN intervals differing by more than 50 milli seconds in the entire recording	numbers	Parasympathetic dominance
	pNN50	proportion of NN50 divided by the total number of NN intervals	percentage %	Parasympathetic dominance

	Variables	Description	Units	Dominance
FREQUENCY DOMAIN	LF power	Power in low frequency range	Milliseconds square (ms^2)	Parasympathetic dominance
	HF power	Power in high frequency range	Milliseconds square (ms^2)	Parasympathetic dominance
	LF/HF Ratio	Ratio LF [ms^2]/HF [ms^2]	percentage %	Sympathetic dominance

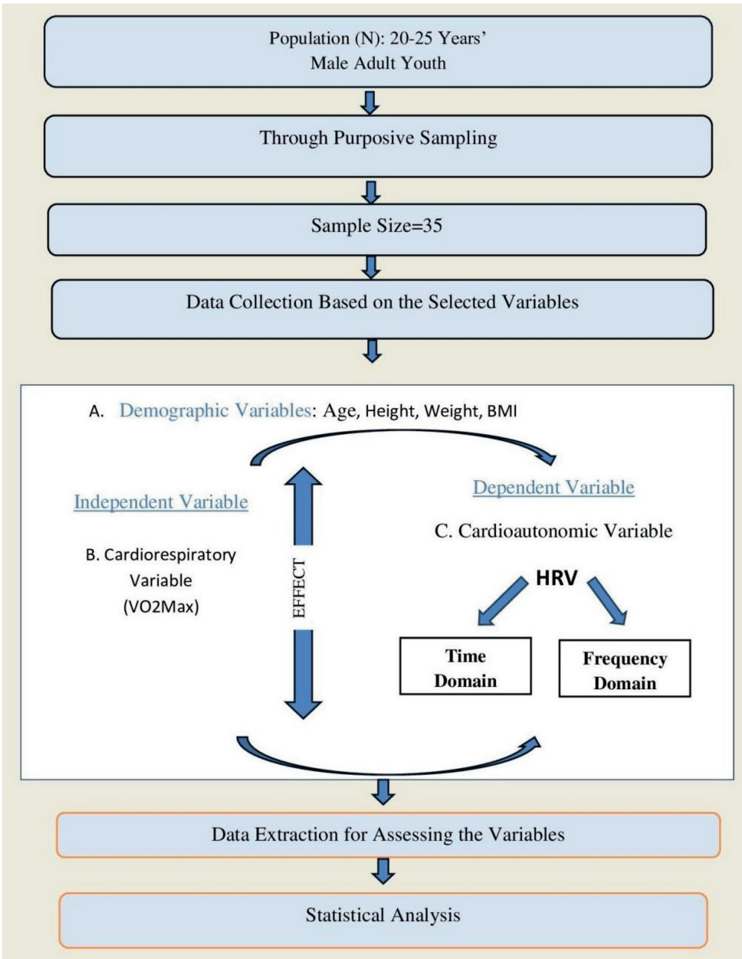

Figure 1. Flowchart of Trial Profile

Statistical procedure

Statistical analyses were performed using SPSS software (version 22.0). The normality of data distribution was assessed using the Shapiro–Wilk test, which confirmed that the data were normally distributed. Descriptive statistics were expressed as mean ± standard deviation. Inferential analysis was conducted using Pearson's product–moment correlation coefficient to examine the relationships between VO_2 max (independent variable) and selected heart rate variability (HRV) indices (RMSSD, SDNN, NN50, pNN50, LF, HF, and LF/HF ratio) as dependent variables. To control for the increased risk of Type I error associated with multiple comparisons, Bonferroni correction was applied to adjust the significance threshold. The level of statistical significance was set at $p \leq 0.05$.

Results

The demographic characteristics of the 35 participants are presented in Table 4. Table 5 summarizes the Pearson correlation coefficients between VO_2max and selected HRV indices, including adjustments for multiple comparisons using the Bonferroni correction. In the time- domain analysis (Figure 2), VO_2 max showed a weak to moderate positive correlation with RMSSD (r=0.340, p=0.045). However, this correlation did not remain significant after applying the Bonferroni correction (adjusted α=0.007). Similarly, the other time-domain indices SDNN (r=0.177, p=0.309), NN50 (r=–0.340, p=0.846), and pNN50 (r=0.198, p=0.254) were not significantly correlated with VO_2 max, both before and after the Bonferroni adjustment. In contrast, in the frequency-domain analysis (Figure 3), VO_2 max showed significant relationships with several HRV indices even after Bonferroni correction. HF power was positively correlated with VO_2 max (r=0.450, p=0.006), indicating that higher aerobic fitness was associated with higher parasympathetic activity. LF power, on the other hand, was negatively correlated with VO_2 max (r=–0.450, p=0.006), indicating that higher VO_2 max was associated with lower sympathetic modulation. Similarly, LF/HF ratio (r=–0.449, p=0.006) was negatively correlated with VO_2 max indicates a shift towards a more balanced or parasympathetic-dominant state, suggesting improved stress recovery and relaxation. All these correlations remained statistically significant even after applying the Bonferroni-adjusted significance threshold of 0.007.

Table 4. Demographic characteristics of participants.

Variables	Mean±SD (n=35)
Age(Years)	23.89±1.64
Height(cm)	169.71±5.52
Weight(kg)	63.94±7.54
BMI	22.09±3.87

Table 5. Descriptive and correlational statistics between VO2 max and selected HRV indices with Bonferroni adjustment.

Independent Variable	Dependent Variable	Mean ± SD	Correlation coefficient (r)	p- value	Bonferroni-adjusted α	Significant
VO2 max (ml/kg/min)	SDNN(ms)	46.94±11	.177	.309	0.007	No
	RMSSD(ms)	49.10±12	.340	.045	0.007	No
	NN50(beats)	199.08±160.48	-.034	.846	0.007	No
	pNN50(%)	26.67±16.69	.198	.254	0.007	No
	LF Power(n.u)	47.19±13.48	-.450	.006	0.007	Yes
	HF Power(n.u)	52.67±13.42	.450	.006	0.007	Yes
	LF/HF	1.04±0.64	-.449	.006	0.007	Yes

Abbreviations: SDNN: standard deviation of the normal-to-normal sinus node-initiated R-R intervals; RMSSD: root mean square of successive R-R interval differences; NN50: Adjacent NN intervals differing by more than 50 milli seconds in the entire recording; pNN50: Proportion of differences in consecutive N-N intervals that are longer than 50 ms; LF: Low frequency; HF: High frequency; LF/HF: Low frequency by high frequency ratio; SD: standard deviation.

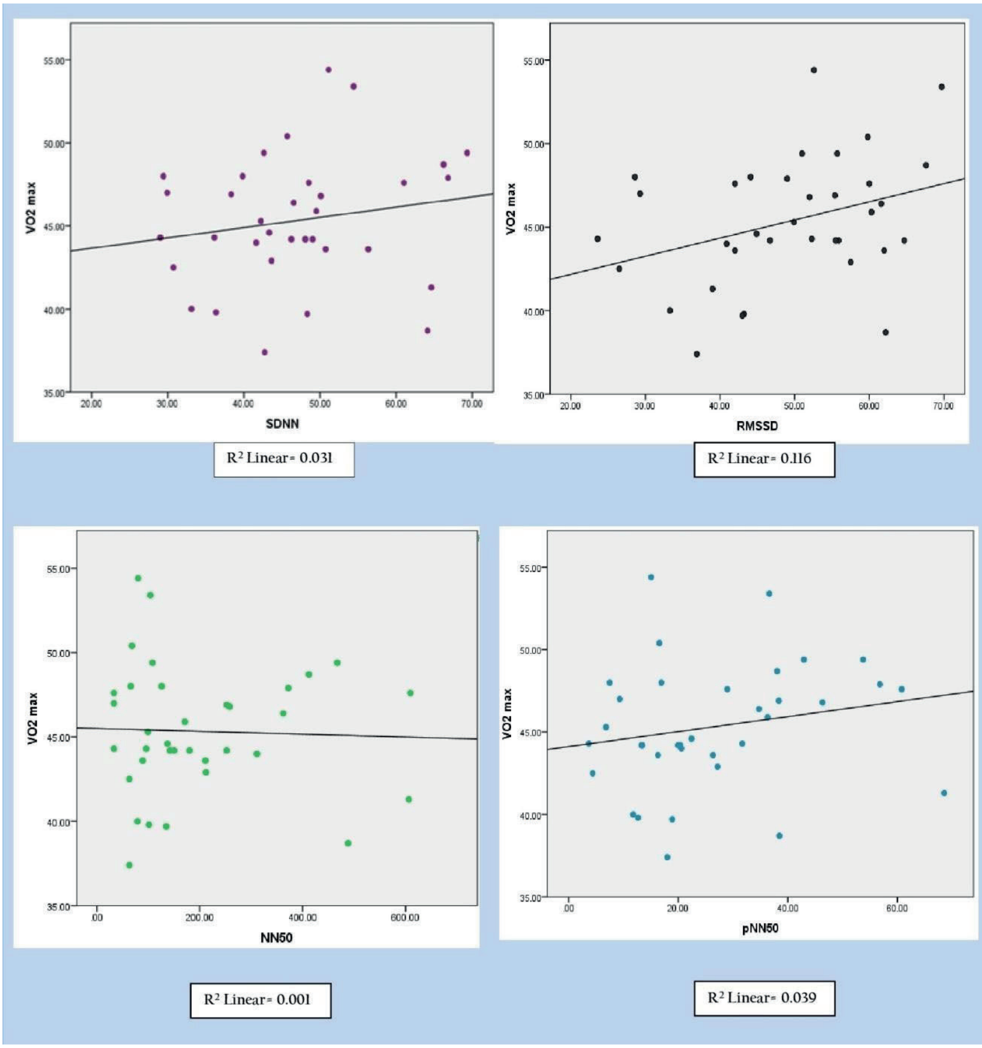

Figure 2. Scatter plot illustrating the correlation among VO2max and Time domain measures of HRV

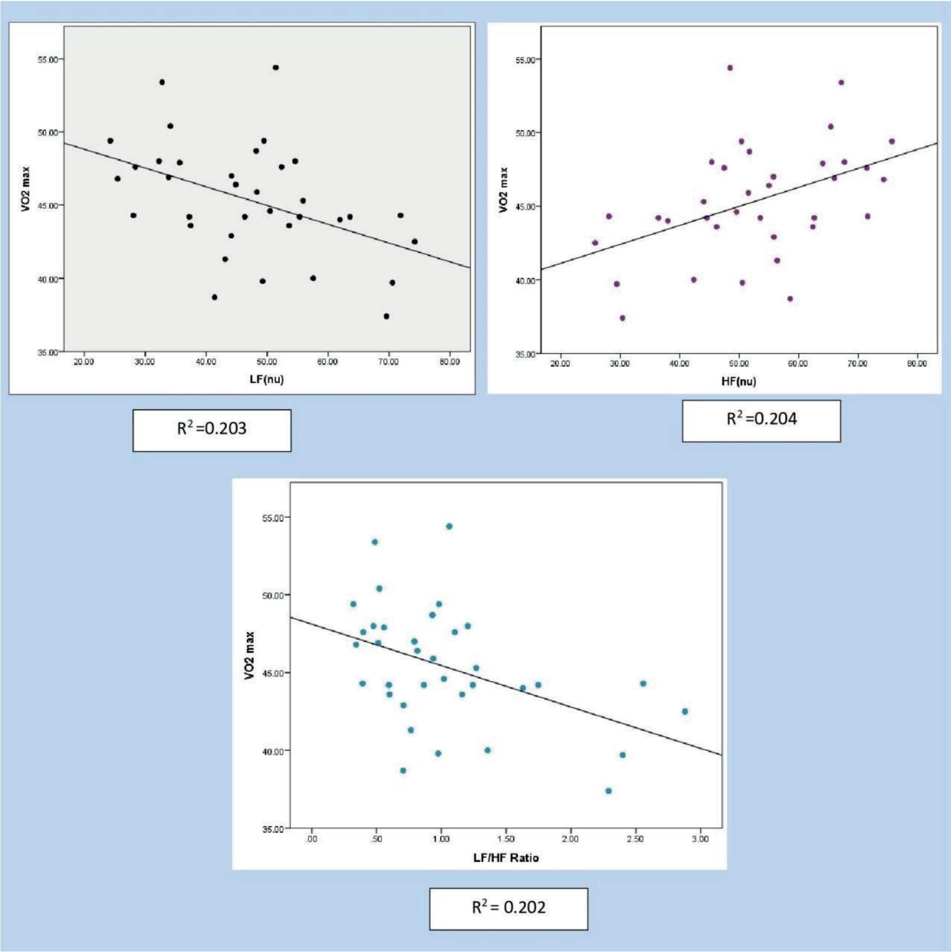

Figure 3. Scatter plot illustrating the correlation among VO2max and Frequency domain measures of HRV

Discussion

HRV time domain and frequency domain indices are related to cardiac electrophysiology and autonomic regulation, are indicative of healthier responses as the existing literature claim that regular physical activity has been associated with increased vagal modulation and reduced sympathetic activity (Swain & Franklin, 2005). This vagal modulation conforms to superior exercise capacity by exercise training which hails usually low resting heart rate. A longitudinal study conducted with a group of untrained young individuals also supported these findings, showing that 6 weeks of aerobic exercise training lowered resting heart rate and enhanced cardiac vagal tone, evaluated through HRV analysis (Al-Ani et al., 1996). Studies conducted on athletes (Daanen et al., 2012; Mann et al., 2014), patients with heart failure (Myers et al., 2007; Pearson & Smart, 2017; Streuber et al., 2006), and patients with Type 2 diabetes (Bhati et al., 2017) have demonstrated that exercise training accelerates heart rate recovery, indicative of enhanced cardiac vagal function. The present study intended here to identify any correlation between aerobic fitness and HRV indices as the available literature has already found a positive correlation between superior exercise and training module with increased aerobic fitness (Wilmore & Knuttgen, 2003). More recent studies also reiterated similar aligned results as the present study. Zaki et al., 2024, showed similar results as of the present study on type two diabetic patients where a positive significant correlation was noted between VO2 max and RMSSD (Zaki et al., 2024). The relationship between the LF/HF ratio and VO2max in another study by Daniela et al., 2022, further supported the results of the present study and the overall notion that higher levels of fitness are associated with a more favourable autonomic balance (Daniela et al., 2022). A weak to moderate positive correlation between RMSSD and VO2max was noted in the present study, also suggested that higher cardiorespiratory fitness levels are associated with enhanced parasympathetic modulation as seen (Cabral et al., 2019). Our findings align with an earlier study by Camillo et al., 2011, which demonstrated that a 12-week high-intensity training regimen incorporating both endurance and strength exercises could enhance HRV measures, including RMSSD, HF and LF/HF (Camillo et al., 2011). Bor ghi-Silva et al., 2009 observed aerobic training program improved some of the time and frequency domain HRV indices (Borghi-Silva et al., 2009). While other time domain aspects SDNN, NN50, pNN50 found a scrawny relation with aerobic fitness. In frequency domains of HRV the researchers found positive significant relation of HF (n.u) with aerobic fitness tend to have stronger parasympathetic modulation of heart rate. This is a marker of good cardiovascular health and recovery capacity whereassignificant negative correlation was found between this resting LF/HF ratio and aerobic fitness which is associated with lower sympathetic dominance and greater parasympathetic activity. As the LF/HF ratio is often implied to be an index of the balance between sympathetic and parasympathetic system henceforth a higher ratio of LF/HF indicates a relative pre dominance of sympathetic activity unlike a lower ratio of LF/HF demonstrated improved cardio-vagal tone. These findings are consistent with research of Gilder & Ramsbottom, 2008 reported that aerobic exercise improves the resting ANS by reducing the sympathetic effect on the heart and boosting the parasympathetic influences (Gilder & Ramsbottom, 2008), similarly Tekin et al., 2025 was found the same result (Tekin et al., 2025). The mechanism of impact of aerobic fitness on cardio autonomic function which has been reflected in the results of the present study, shown in Figure 4.

This research study has established positive and significant connections between aerobic fitness and cardiac autonomic modulation associated with HRV. The result of present study also showed that individuals with higher aerobic fitness levels tend to exhibit greater heart rate variability. VO2 max, a marker of cardiovascular endurance, appears to be linked with improved autonomic nervous system function, as reflected by higher HRV. This relationship indicates that individuals with better cardiovascular fitness may experience enhanced parasympathetic modulation, contributing to improved recovery and stress resilience. These findings emphasize the importance of aerobic fitness not only for physical performance but also for optimal autonomic regulation and overall cardiovascular health.

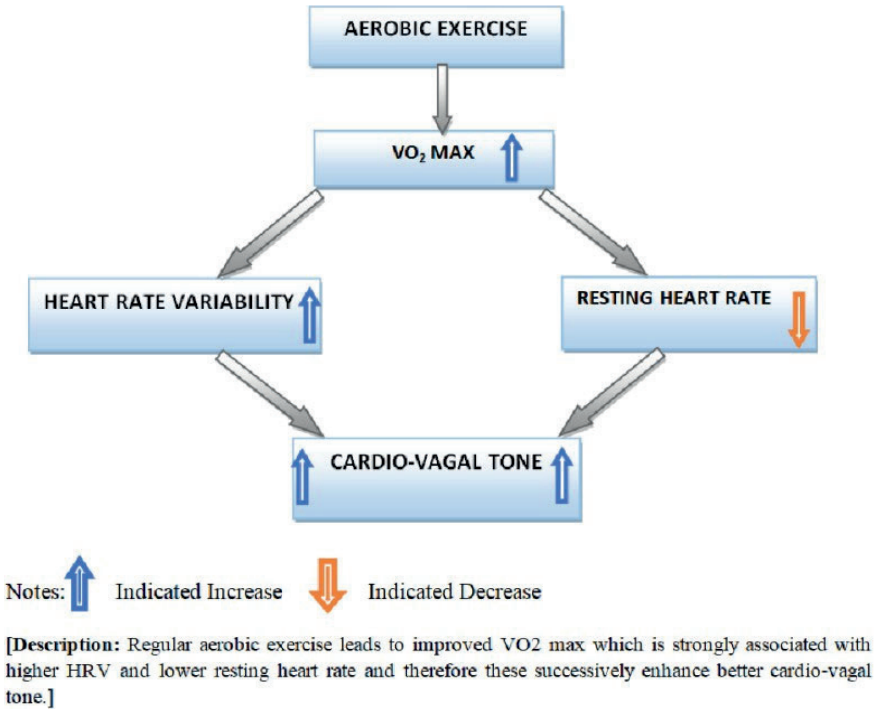

Figure 4. Mechanism of Cardio-vagal adaptation through Aerobic Exercise.

Conclusion

This study identified a significant correlation between VO_2 max and some of the selected HRV indices, supporting the notion that regular aerobic exercise or physical activity enhances cardio-vagal regulation and promotes a more balanced cardiac autonomic function. The current investigation focused only on male young adult participants who engaged in regular exercise and had a minimum training experience of four years. Considering the constraints of this study, involving elite athletes from various sports backgrounds, a larger sample size, and representation from both genders, along with long-term HRV monitoring, may yield improved outcomes and enhance understanding in future research.

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Cite this article as: Paul, P., Roy, D., Bhattacharya, P., Chatterjee, S. (2026). Effect of Aerobic Fitness on Cardio-Vagal Adaptation. *Central European Journal of Sport Sciences and Medicine*, *1*(53), 87–98, https://doi.org/10.18276/cej.2026.1-08