Association between heart rate variability and cardiorespiratory fitness in individuals with type 2 diabetes mellitus: A cross-sectional study

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ABSTRACT

Background: Type 2 Diabetes Mellitus (T2DM) is associated with cardiovascular risk, which is partly due to autonomic dysfunction and decreased cardiorespiratory fitness. This study examines the relationship between heart rate variability (HRV) and maximal oxygen uptake (VO$_{2\text{max}}$) in T2DM patients to understand their interconnected impacts on autonomic and aerobic functions. Methods: In a cross-sectional study, 77 T2DM patients underwent HRV and VO$_{2\text{max}}$ assessments using standard protocols. HRV metrics were analysed in conjunction with VO$_{2\text{max}}$, measured through direct breath-by-breath analysis. Pearson’s correlation coefficient was used to investigate the relationships between HRV indices and VO$_{2\text{max}}$. Results: VO$_{2\text{max}}$ showed strong positive correlations with RMSSD (r = 0.89, p < .001), HF (r = 0.54, p < .001), and pNN50% (r = 0.52, p < .001), indicating higher parasympathetic activity with improved cardiorespiratory fitness. Negative correlations with LF (r = -0.60, p < .001) and the LF/HF ratio (r = -0.39, p < .001) suggested that better fitness levels lead to sympathetic withdrawal and a more favourable autonomic balance. Moderate positive correlations with SDNN (r = 0.46, p < .001) and TP (r = 0.58, p < .001) further suggested that overall autonomic modulation is enhanced with increased cardiorespiratory fitness. Conclusion: This study substantiates a significant correlation between HRV and VO$_{2\text{max}}$ in individuals with T2DM, highlighting the intricate relationship between autonomic function and aerobic capacity. These findings suggest that enhancing cardiorespiratory fitness may improve autonomic balance, offering potential avenues for mitigating cardiovascular risk in the T2DM population.

Keywords: Sport medicine, Cardiac autonomic modulation, Exercise testing, Parasympathetic, Sympathetic.

INTRODUCTION

Type 2 Diabetes Mellitus (T2DM) represents a significant global health concern, marked by its complex pathophysiology and an elevated risk of cardiovascular complications (Galicia-Garcia et al., 2020). This metabolic disorder affects millions worldwide, underscoring the importance of understanding and managing its multifaceted impact on health (Zheng et al., 2018). Among the various factors influencing the cardiovascular sequelae of T2DM, the autonomic control of the heart, as reflected by heart rate variability (HRV), stands out for its predictive value regarding cardiovascular health and outcomes (Benichou et al., 2018). HRV, a non-invasive marker of autonomic nervous system functionality, reveals the heart's ability to respond to varying physiological conditions, highlighting its relevance in the context of diabetes management (Shaffer & Ginsberg, 2017).

Concurrently, cardiorespiratory fitness (CRF), often quantified by maximal oxygen uptake (VO2max), is a robust predictor of cardiovascular and all-cause mortality, underlining its crucial role in assessing overall health and the risk of chronic diseases (Gonzales et al., 2021). The significance of VO2max extends beyond its representation of physical fitness; it encapsulates the efficiency of the cardiovascular, respiratory, and muscular systems to supply and utilize oxygen during sustained physical activity (Gim & Choi, 2016). This efficiency is paramount, especially in the context of T2DM, where decreased CRF is a common finding (Regensteiner et al., 1995; Tadic et al., 2021). Individuals with T2DM typically exhibit lower VO2max values compared to their nondiabetic counterparts, a disparity that not only marks impaired physical fitness but also signifies an elevated risk of cardiovascular complications and reduced quality of life (Cai et al., 2023).

Given that T2DM is characterized by insulin resistance and metabolic dysregulation (Galicia-Garcia et al., 2020), which have been associated with reduced HRV and impaired CRF. For instance, a study by Larsen et al. (2012) demonstrated that CRF is highly correlated with insulin sensitivity and secretion in a healthy population, suggesting the potential for similar associations in T2DM (Larsen et al., 2012). Further, investigations such as the 23-year cohort study by Zaccardi et al. (2015) have underscored the inverse relationship between CRF and T2DM risk, independent of other factors (Zaccardi et al., 2015). The relationship between HRV and cardiovascular health indicators, such as endothelial function and physical fitness, especially in T2DM, further elucidates the intertwined nature of these metrics (Picard et al., 2021; Tuttolomondo et al., 2021). Additionally, Kadoglou et al. (2009) found that cardiorespiratory capacity is associated with a favourable cardiovascular risk profile in T2DM patients, offering a further insight into the potential benefits of improving HRV and VO2max in this demographic (Kadoglou et al., 2009).

While both HRV and VO2max have independently demonstrated associations with cardiovascular disease risk and mortality in various populations, their interrelationship, particularly in the context of T2DM, remains relatively unexplored. Existing literature predominantly examines these parameters in isolation or in non-diabetic population (Benichou et al., 2018; Granero-Gallegos et al., 2020; Grant et al., 2013; Leite et al., 2009). This study uniquely focuses on individuals with T2DM, a group in which these relationships are less understood and potentially more complex due to the interplay of metabolic, cardiovascular, and autonomic factors. Therefore, the aim of this study is to determine the correlation of HRV metrics align with VO2max, in a T2DM population. We hypothesize that there exists a significant correlation between HRV metrics and VO2max in individuals with T2DM, suggesting an intertwined relationship between autonomic function and aerobic capacity in this population.
METHODS

Sample size calculation
The sample size of this study was determined using G*Power 3.1.9.4, implementing a bivariate normal model for correlation analysis. The power analysis was conducted a priori to compute the necessary sample size, given alpha, power, and the anticipated effect size. We selected a one-tailed hypothesis test with an alpha error probability of 0.05, and we aimed for a power of 0.85, indicating a 15% probability of a Type II error. The effect size for the expected correlation (ρ H1) was set at 0.3, reflecting a medium effect size following Cohen’s conventions (Cohen, 2013). Based on these parameters, the analysis yielded a sample size of 77 participants.

Sampling
A cross-sectional study was conducted on 77 patients with T2DM at Centre for Physiotherapy and Rehabilitation Sciences, Jamia Millia Islamia, New Delhi, India. The participant recruitment was performed from outpatient clinics of MA Ansari Health Centre and the Physiotherapy clinic of Centre for Physiotherapy and Rehabilitation Sciences, Jamia Millia Islamia, New Delhi, India by convenience sampling. Participants included in the study were required to have been diagnosed with T2DM for a minimum of one year and be between the ages of 30 and 70 years. Exclusion criteria included severe arrhythmias, pregnancy, breastfeeding, other metabolic disorders, significant comorbidities, contraindications for exercise testing, and acute complications related to T2DM (Ferguson, 2014; Tang et al., 2013).

Procedure
The cross-sectional study was conducted from August 2023 to January 2024, adhering to the STROBE (Strengthening the Reporting of Observational Studies in Epidemiology) guidelines for reporting cross-sectional research (Cuschieri, 2019). All participants have provided their informed consent prior participation in this study. The research followed the ethical guidelines set forth in the Declaration of Helsinki (Ashcroft, 2008). The eligibility criteria were evaluated by medical professionals at Dr. M. A Ansari Health Centre, Jamia Millia Islamia, New Delhi, India. On the first day, eligible participants underwent an assessment of general demographic details, including age, height, weight, gender, and body mass index (BMI), along with clinical parameters such as the duration of diabetes, resting heart rate, systolic blood pressure (SBP), and diastolic blood pressure (DBP). Instructions for the procedures on subsequent days were provided at this time. On the second day of the study, participants underwent assessments for HRV and VO\textsubscript{2max}. Resting HRV measurements were taken initially, followed by the evaluation of VO\textsubscript{2max}. Blood pressure and body mass index assessment
The SBP and DBP measurements were taken using a conventional sphygmomanometer. Participants were instructed to avoid caffeine, exercise, and smoking for 30 minutes before their assessment. They were asked to sit calmly for 5 minutes. After a 10-minute rest period in the seated position, blood pressure readings were obtained with the sphygmomanometer. The cuff was applied to either the right or left arm, and the initial blood pressure reading was recorded. A follow-up reading was conducted 2 minutes later on the same arm to ensure accuracy. The final blood pressure reading was determined by taking the average of two consecutive stable measurements (Muntner et al., 2019). Subsequently, after a brief rest interval of 1-2 minutes, a recording was taken from the opposite arm as well. In cases where discrepancies were observed between the measurements from both arms, the arm with the higher reading was selected for the purpose of analysis (Muntner et al., 2019).
For the assessment of BMI, height and weight measurements were accurately obtained using standard medical scales and a stadiometer. Participants were requested to remove heavy clothing and shoes for precise measurement. BMI was then calculated by dividing the participant's weight in kilograms by the square of their height in meters (kg/m²) (Weir & Jan, 2019).

**Assessment of resting heart rate variability**

The evaluation of HRV for all participants was scheduled between 9:00 AM and 12:00 PM, after ensuring a fasting period of at least 2 hours post-meal. This specific timing was selected to reduce the impact of circadian rhythms on autonomic function and to prevent any potential disturbances in test outcomes caused by recent food consumption (Bhati & Hussain, 2021). Participants were advised to avoid medications that could alter autonomic nervous system activity for at least 24 hours before undergoing tests for autonomic function and blood pressure. This guideline follows the recommendations issued by the Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology in 1996 ("Heart rate variability. Standards of measurement, physiological interpretation, and clinical use. Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology," 1996).

After resting for at least 15 minutes in a supine position within an environment maintained at 24°C, an electrocardiogram (ECG) was captured for a duration of 10 minutes using a standard lead II setup. Analysis focused on the final 5 minutes of the ECG recording to examine both time and frequency domain aspects of HRV. This analysis was performed utilizing LabChart software version 7.3.7 (Power Lab 8 SP, AD Instruments, Australia) to process the data. Careful examination was undertaken to identify and interpolate ectopic beats, ensuring they did not exceed 10% of all beats for a consistent dataset. The analysis employed Fast Fourier Transform for power spectral analysis to delineate signal power into its frequency components, applying a lowpass filter with a 40 Hz cut-off to the data. Evaluated metrics included Total Power (TP), Low Frequency (LF) in millisecond square (ms²), High Frequency (HF) in ms², and the LF/HF ratio. Time domain measures such as the standard deviation of all normal-to-normal intervals (SDNN), the root mean square of successive differences between normal heartbeats (RMSSD), and the percentage of successive R-R intervals varying by more than 50 ms (pNN50%) were also assessed ("Heart rate variability. Standards of measurement, physiological interpretation, and clinical use. Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology," 1996).

**Assessment of cardiorespiratory fitness**

In this study, VO₂max measurements were obtained through a direct breath-by-breath analysis technique, which is critical for evaluating CRF. This process involved the use of an open circuit spirometry system. Participants inhaled and exhaled through a mouthpiece connected to a gas analysis unit (Model: ML206), allowing for the precise capture of respiratory data. This data was then analysed with the Metabolic Module within LabChart Software by AD Instruments, ensuring the integrity and accuracy of VO₂max calculations. This technique aligns with established protocols for accurately determining VO₂max, underscoring the efficacy of breath-by-breath analysis in measuring aerobic fitness levels (Beltz et al., 2016).

The study exclusively implemented the modified Bruce protocol during the graded exercise test on a treadmill. Unlike the traditional Bruce protocol, this modified approach starts at a lower intensity, progressively escalating through stages that last 3 minutes each. This gradual increase from a gentle walk to more intense levels of speed and incline is specifically designed to accommodate a wide range of participant fitness levels (Trabulo et al., 1994). Such customization ensures that all participants can safely achieve their maximal exertion. Continuous monitoring of heart rate, oxygen uptake, and expired gases was conducted throughout
the test. This provided real-time feedback necessary for the accurate calculation of VO$_{2\text{max}}$, facilitating a thorough assessment of each participant's cardiopulmonary fitness.

**Statistical analysis**

The statistical analysis in this study was conducted with the aid of SPSS version 21 (SPSS Inc., Chicago, Illinois) and MedCalc Statistical Software version 19.2.6 (MedCalc Software bv, Ostend, Belgium). Data are presented as mean and standard deviation unless otherwise specified. The normality of data distribution was assessed using the Shapiro-Wilk test. For variables not normally distributed, log transformation was applied to achieve normality. The correlation between HRV indices and VO$_{2\text{max}}$ was analysed using Pearson's correlation coefficient to determine the strength and direction of the association, which provided insights into both the strength and direction of the association. Interpretation of Pearson's correlation coefficient values was based on the established ranges: values from -0.19 to 0.19 indicate a very weak correlation, 0.20 to 0.39 (or -0.20 to -0.39) a weak correlation, 0.40 to 0.59 (or -0.40 to -0.59) a moderate correlation, 0.60 to 0.79 (or -0.60 to -0.79) a strong correlation, and 0.80 to 1.0 (or -0.80 to -1.0) a very strong correlation. These value ranges are used to quantify the linear relationship between two variables, where the closer the value is to ±1, the stronger the relationship (Akoglu, 2018).

**RESULTS**

The demographic and clinical characteristics of the 77 participants with T2DM are illustrated in Table 1, revealed an average age of 46.8 ± 7.7 years, with a male predominance (46 males and 31 females). The HRV indices demonstrated significant correlations with VO$_{2\text{max}}$ levels among participants (Table 2). Specifically, RMSSD, indicative of parasympathetic activity, showed the strongest positive correlation with VO$_{2\text{max}}$ ($r = 0.89$, $p < .001$), indicating a strong relationship (Figure 1). On the other hand, LF, a marker of sympathetic activity, was negatively correlated with VO$_{2\text{max}}$ ($r = -0.60$, $p < .001$), suggesting that improvements in cardiorespiratory fitness are associated with reduced sympathetic dominance (Figure 2). The LF/HF ratio, which assesses the balance between sympathetic and parasympathetic activity, also negatively correlated with VO$_{2\text{max}}$ ($r = -0.39$, $p < .001$), indicating that higher fitness levels are associated with a more favourable autonomic balance (Figure 3).

<table>
<thead>
<tr>
<th>Variables</th>
<th>Mean ± SD (n = 77)</th>
</tr>
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<tbody>
<tr>
<td>Age (years)</td>
<td>46.8 ± 7.7</td>
</tr>
<tr>
<td>Sex (M/F)</td>
<td>46/31</td>
</tr>
<tr>
<td>BMI (kg/m$^2$)</td>
<td>33.5 ± 4.7</td>
</tr>
<tr>
<td>T2DM duration (years)</td>
<td>6.9 ± 0.5</td>
</tr>
<tr>
<td>SBP (mmHg)</td>
<td>131.5 ± 13.3</td>
</tr>
<tr>
<td>DBP (mmHg)</td>
<td>83.3 ± 4.8</td>
</tr>
<tr>
<td>HR$_{\text{rest}}$ (beats/min)</td>
<td>81.2 ± 11</td>
</tr>
<tr>
<td>FBG (mg/dl)</td>
<td>110 ± 5</td>
</tr>
<tr>
<td>PPBG (mg/dl)</td>
<td>159.7 ± 6.8</td>
</tr>
<tr>
<td>HbA1c (%)</td>
<td>6.2 ± 0.3</td>
</tr>
<tr>
<td>VO$_{2\text{max}}$ (ml/kg/min)</td>
<td>26.6 ± 6.4</td>
</tr>
</tbody>
</table>

Abbreviations: M: male; F: female; BMI: body mass index; HR$_{\text{rest}}$: resting heart rate; SBP: systolic blood pressure; DBP: diastolic blood pressure; FBG: fasting blood glucose; PPBG: post-prandial blood glucose; HbA1c: glycosylated haemoglobin; VO$_{2\text{max}}$: volume of maximum oxygen consumption.
Table 2. Descriptives, and correlation statistics of HRV indices with \( \text{VO}_{2\text{max}} \).

<table>
<thead>
<tr>
<th>Variables</th>
<th>Mean ± SD</th>
<th>Correlation coefficient (r)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>SDNN (ms)</td>
<td>50.1 ± 1.3</td>
<td>0.46</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>pNN50 (%)</td>
<td>32.4 ± 13.7</td>
<td>0.52</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>RMSSD (ms)</td>
<td>59.6 ± 9</td>
<td>0.89</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>LF (ms(^2))</td>
<td>293 ± 123.2</td>
<td>-0.60</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>HF (ms(^2))</td>
<td>406.6 ± 159.5</td>
<td>0.54</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>TP (ms(^2))</td>
<td>952.5 ± 129</td>
<td>0.58</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>LF/HF</td>
<td>1.1 ± 1</td>
<td>-0.39</td>
<td>&lt;.001</td>
</tr>
</tbody>
</table>

Abbreviations: SDNN: standard deviation of the normal-to-normal sinus node-initiated R-R intervals; pNN50%: Proportion of differences in consecutive N-N intervals that are longer than 50 ms; RMSSD: root mean square of successive R-R interval differences; LF: Low frequency; HF: High frequency; TP: Total power; LF/HF: Low frequency by high frequency ratio; SD: standard deviation.

Figure 1. Scatter plot illustrating the correlation between root mean square of successive differences between normal heartbeats (RMSSD) and \( \text{VO}_{2\text{max}} \) in individuals with Type 2 Diabetes.

Figure 2. Scatter plot illustrating the correlation between Low Frequency (LF) and \( \text{VO}_{2\text{max}} \) in individuals with Type 2 Diabetes.
The indicators of parasympathetic control including HF ($r = 0.54$, $p < .001$) and pNN50\% ($r = 0.52$, $p < .001$) have demonstrated moderate significant correlation (Figure 4 and Figure 5). Additionally, SDNN ($r = 0.46$, $p < .001$) and TP ($r = 0.58$, $p < .001$) were moderately correlated, indicating increase in overall autonomic modulation is associated with better VO$_{2\text{max}}$ level (Figure 6 and Figure 7). These findings highlight the intricate relationship between autonomic function and cardiorespiratory fitness, with significant influence by both sympathetic and parasympathetic influences, alongside overall sympathovagal balance.
DISCUSSION

The present study aimed to elucidate the correlation between cardiac autonomic control, as measured by HRV, and CRF, quantified through VO\textsubscript{2max}, in individuals with T2DM. Our findings reveal a linear relationship between HRV and VO\textsubscript{2max}, significantly influenced by both sympathetic and parasympathetic branches of the autonomic nervous system, as well as the overall sympathovagal balance. This relationship underscores the potential for HRV and VO\textsubscript{2max} as critical markers in assessing cardiovascular risk and fitness levels in a population inherently at higher risk for cardiovascular diseases. Such insights contribute to a deeper understanding of the complex interplay between autonomic control and physical fitness in the context of type 2 diabetes. 
understanding of the autonomic and cardiovascular interplay in T2DM, highlighting the importance of integrated approaches in managing and mitigating cardiovascular risk among these patients.

![Figure 7](image)

**Figure 7.** Scatter plot illustrating the correlation between Total Power (TP) and VO$_{2\text{max}}$ in individuals with Type 2 Diabetes.

The strong positive correlation between RMSSD and VO$_{2\text{max}}$, suggests that higher cardiorespiratory fitness levels are associated with enhanced parasympathetic modulation (Cabral et al., 2019). This is significant because enhanced parasympathetic activity is known to be cardioprotective, reducing the risk of arrhythmias and cardiac events (Shaffer & Ginsberg, 2017). Conversely, the negative correlation between LF and VO$_{2\text{max}}$ underscores the concept that improved fitness levels might lead to a reduction in sympathetic dominance, which is beneficial since excessive sympathetic activity is associated with adverse cardiovascular outcomes (Tuttolomondo et al., 2021). The inverse relationship between the LF/HF ratio and VO$_{2\text{max}}$ further supports the notion that higher levels of fitness are associated with a more favourable autonomic balance, possibly reflecting better cardiovascular health (Daniela et al., 2022). This balance is critical in T2DM patients, where autonomic dysfunction can exacerbate the risk of cardiovascular complications (Tuttolomondo et al., 2021).

Further, the observed moderate positive correlation between SDNN and VO$_{2\text{max}}$ indicates that individuals with higher CRF exhibit greater overall variability in heart rate, suggesting a healthier autonomic nervous system and a lower risk of cardiac complications (Shaffer & Ginsberg, 2017). Similarly, the significant association between pNN50% and VO$_{2\text{max}}$ highlights the role of acute changes in HRV in response to stressors, further emphasizing the benefits of enhanced parasympathetic activity (Picard et al., 2021). Additionally, the relationship between TP and VO$_{2\text{max}}$ underscores the holistic influence of physical fitness on the spectrum of autonomic heart rate modulation. Higher TP values, indicative of overall HRV, suggest that increased fitness levels are associated with an autonomic system capable of a wider range of responses to physiological stimuli, a characteristic deemed protective against cardiovascular morbidity (Granero-Gallegos et al., 2020; Picard et al., 2021).

Previous research has shown similar associations, indicating that individuals with T2DM often exhibit reduced HRV and VO$_{2\text{max}}$, reflecting impaired autonomic regulation and diminished CRF. For instance, Larsen et al.
(2012) found that CRF is highly correlated with insulin sensitivity (Larsen et al., 2012), indicating that improved VO\textsubscript{2max} could have beneficial effects on metabolic control in T2DM. Further, Loimaala et al. (2003) demonstrated that exercise training enhances baroreflex sensitivity and HRV, alongside improvements in VO\textsubscript{2max}, suggesting that physical activity can ameliorate autonomic balance and cardiovascular risk in T2DM patients (Loimaala et al., 2003). Moreover, the metabolic improvements associated with higher VO\textsubscript{2max}, such as better glucose control and reduced insulin resistance, are particularly beneficial in T2DM, potentially contributing to the observed relationships (Zaki et al., 2023). These metabolic benefits, alongside improved autonomic balance, could explain the strong correlations observed in the study.

**Clinical implications**

From a clinical perspective, these findings emphasize the importance of promoting physical activity and improving CRF as non-pharmacological strategies to mitigate cardiovascular risk in T2DM. Given the modifiable nature of HRV and VO\textsubscript{2max} through lifestyle interventions, these markers could serve as valuable tools in monitoring the effectiveness of interventions aimed at reducing cardiovascular risk in the T2DM population.

**Limitations and future research**

While the study's findings are robust, it is essential to acknowledge its cross-sectional design, which limits the ability to infer causality. Longitudinal studies are needed to ascertain the causal relationships between HRV, VO\textsubscript{2max}, and cardiovascular outcomes in T2DM. Additionally, exploring the impact of specific types of physical activity and their intensity on HRV and VO\textsubscript{2max} could provide more nuanced guidelines for managing T2DM.

**CONCLUSION**

This study demonstrated the significant correlation between HRV metrics and VO\textsubscript{2max}, underscoring the complex interplay between autonomic function and CRF in T2DM. These findings not only advance our understanding of the physiological mechanisms at play but also highlight the potential of incorporating HRV and VO\textsubscript{2max} assessments in the clinical management of T2DM to improve cardiovascular health outcomes.

**AUTHOR CONTRIBUTIONS**

The full manuscript has been read and approved by all authors. Each listed author fulfils the requirements for authorship, and each author attests that the manuscript represents honest work.

**SUPPORTING AGENCIES**

No funding agencies were reported by the authors.

**DISCLOSURE STATEMENT**

No potential conflict of interest were reported by the authors.

**RESEARCH QUALITY AND ETHICS STATEMENT**

Ethical clearance for this study was granted by the Institutional Ethics Committee, Jamia Millia Islamia (Approval number: 24/5/323/JMII/IEC/2021, on July 7, 2021). The research was carried out as part of a larger
project, which has been registered with the Clinical Trial Registry India (CTRI/2021/09/036711, registration date September 21, 2021). In conducting this study, the authors adhered to relevant guidelines from the EQUATOR Network, specifically following the STROBE guideline.

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Correlation Study on HRV and VO$_2$max in Diabetes


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