Research Article

Assessment of Cardiac Autonomic Neuropathy Using Time-Domain HRV: A Case-Control Study in Type 2 Diabetes

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ABSTRACT

Background: Cardiac autonomic neuropathy (CAN) is a clinically significant yet frequently underrecognized complication of type 2 diabetes mellitus (T2DM). Heart rate variability (HRV) analysis provides a non-invasive method for evaluating cardiac autonomic function. This study aimed to assess time-domain HRV parameters in asymptomatic patients with T2DM compared to healthy controls.
Methods: In this hospital-based case-control study, 50 patients with T2DM and 50 age-matched healthy controls (40–60 years) were enrolled following institutional ethics approval. Short-term (5-minute) ECG recordings were obtained under standardized conditions. Time-domain HRV indices - mean RR interval, heart rate, SDNN, RMSSD, pNN50%, RRΔ index, and TINN were analyzed. Group comparisons were performed using independent samples t-test. Effect sizes (Cohen's d) and 95% confidence intervals (CIs) were calculated, and Bonferroni correction was applied for multiple comparisons.
Results: Compared with controls, patients with T2DM exhibited significantly reduced SDNN (mean difference −6.60 ms; 95% CI: −10.4 to −2.8), RMSSD (−10.39 ms; 95% CI: −15.6 to −5.1), pNN50% (−4.69%; 95% CI: −5.6 to −3.7), and TINN (−155.1 ms; 95% CI: −210.4 to −99.8) (adjusted $p < 0.01$). The T2DM group also demonstrated a lower mean RR interval and higher resting heart rate. Effect sizes ranged from moderate to large (Cohen's d = 0.6–1.1).
Conclusion: Time-domain HRV parameters were significantly reduced in asymptomatic individuals with T2DM, suggesting early subclinical cardiac autonomic dysfunction. Short-term HRV assessment may serve as a practical screening tool for early detection of CAN.

Keywords: Heart rate variability, Type 2 diabetes mellitus, Cardiac autonomic neuropathy, Time-domain analysis.

1. INTRODUCTION

Type 2 diabetes mellitus (T2DM) is a major global public health problem with rapidly increasing prevalence [1]. Chronic hyperglycemia contributes to microvascular and macrovascular complications including neuropathy and cardiovascular disease [1]. Cardiac autonomic neuropathy (CAN) is a serious complication associated with increased cardiovascular morbidity and mortality [2,5]. CAN results from damage to autonomic nerve fibers supplying the heart, leading to impaired heart rate control and abnormal vascular dynamics [6]. Early stages are often asymptomatic but may progress to resting tachycardia, silent myocardial ischemia, and increased risk of sudden cardiac death [5]. Therefore, early detection of subclinical autonomic dysfunction is clinically important. Heart rate variability (HRV) reflects beat-to-beat variations in RR intervals and serves as a surrogate marker of autonomic nervous system (ANS) modulation [3]. HRV can be assessed using time-domain, frequency-domain, and non-linear methods [3,4]. Time-domain analysis involves statistical evaluation of normal-to-normal (NN) intervals and is recommended for

short-term recordings in clinical research [3]. Parameters such as SDNN reflect overall autonomic modulation, while RMSSD and pNN50% predominantly represent parasympathetic activity [3,4]. Reduced HRV has been consistently reported in patients with T2DM and is considered an early marker of CAN [2,7]. Although frequency-domain analysis provides additional insight into sympathetic–parasympathetic balance, time-domain metrics were selected in this study because they are simple, reproducible, and clinically validated for short-term recordings [3]. However, HRV interpretation may be influenced by age, blood pressure, medications, and metabolic status [8], and does not replace gold-standard autonomic reflex testing [6]. The present study aimed to assess time-domain HRV parameters in asymptomatic T2DM patients compared to healthy controls and evaluate their role in early detection of CAN.

2. Materials and Methods:

Study Design

This hospital-based case-control study was conducted at Yenepoya Medical College Hospital, Mangalore, after approval from the Institutional Ethics Committee. The study adhered to ethical principles for medical research involving human subjects [9].

Participants

A total of 100 participants were included:

- 50 diagnosed T2DM patients (cases)
- 50 apparently healthy controls

Controls were frequency-matched for age (40-60 years). The selected age range minimized age-related confounding, as HRV declines with advancing age [3].

Inclusion Criteria

- Diagnosed T2DM (40–60 years)
- Healthy controls (40–60 years)

Exclusion Criteria

- Declined participation or unable to provide consent
- Pregnancy
- Hypertension or cardiovascular disease
- Atrial fibrillation
- Known macrovascular complications
- Clinically overt neuropathy
- Smoking or alcohol use
- Medications affecting autonomic function

Clinical Characteristics of Diabetic Subjects

Duration of diabetes and treatment regimen (oral hypoglycemic agents and/or insulin therapy) were recorded. Subjects with macrovascular complications were excluded. HbA1c was not recorded in this study.

Sample Size Calculation

Sample size estimation was based on previously reported SDNN differences between diabetic and non-diabetic subjects [7]. Assuming an effect size of 0.6, $\alpha = 0.05$, and power of 80%, at least 45 participants per group were required.

HRV Recording Protocol

- Recording time: 9:00 am - 11:00 am
- Posture: Seated
- Rest period: 10 minutes prior
- Duration: 5-minute ECG (Lead II)
- Environment: Quiet room, 22–24°C
- Caffeine/exercise restriction: 12 hours

Ectopic beats and artifacts were detected and corrected using cubic spline interpolation as recommended in HRV analysis standards [3]. Only NN intervals were analyzed.

HRV Parameters

- SDNN: Standard deviation of NN intervals (overall variability)
- RMSSD: Parasympathetic activity marker
- pNN50%: Percentage of successive NN intervals differing by >50 ms
- TINN: Triangular interpolation of NN histogram
- RRΔ index: Mean successive RR interval differences

3. Statistical Analysis

Data were analyzed using SPSS software (IBM Corp.). Normality was assessed using the Shapiro–Wilk test. Independent samples t-test was used for normally distributed variables.

Given multiple HRV comparisons, Bonferroni correction was applied. Effect sizes (Cohen's d) were calculated.

A p-value < 0.05 was considered statistically significant.

4. Results

Patients with T2DM demonstrated significantly reduced SDNN, RMSSD, pNN50%, RR interval, RRΔ index, and TINN compared to controls. Resting heart rate was significantly higher in the diabetic group.

These findings indicate reduced parasympathetic modulation and relative sympathetic predominance in T2DM.

Table 1: Comparison of anthropometric parameters & blood pressure between diabetics and controls.

Parameters	Cases (Mean ± SD)	Controls (Mean ± SD)	p- value
Age (yrs)	51.88 ± 5.69	49.16 ± 5.33	0.015*
Height (m)	1.64 ± 0.08	1.63 ± 0.06	0.25
Weight (kg)	64.24 ± 8.14	64.12 ± 6.12	0.934
BMI (kg/m^2)	23.84 ± 1.94	24.32 ± 1.95	0.22
Systolic BP (mmHg)	126.44 ± 13.59	128.36 ± 15.73	0.515
Diastolic BP (mm Hg)	79.80 ± 8.02	82.20 ± 8.18	0.142

* Significant (p-value<0.05)

Table 2: Comparison of time domain analysis of heart rate variability between diabetics and controls.

Parameters	Cases (Mean ± SD)	Controls (Mean ± SD)	p- value
SDNN (ms)	60.12 ± 10.06	66.72 ± 10.49	0.002*
RMSSD (ms)	20.22 ± 13.88	30.61 ± 12.55	0.001*
RR (ms)	820.40 ± 214.10	941.50 ± 137.14	0.001*
Heart Rate (bpm)	71.44 ± 8.18	66.70 ± 7.86	0.004*
pNN50%	1.63 ± 1.88	6.32 ± 2.76	0.002*
RRΔ Index	0.05 ± 0.01	0.07 ± 0.02	0.001*
TINN (ms)	143.80 ± 101.69	298.94 ± 202.96	0.001*

* Significant (p-value<0.05)

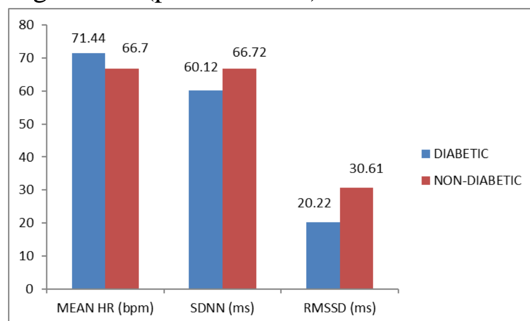

Figure 1: Comparison of time domain analysis (Mean HR, SDNN AND RMSSD) between diabetic and non-diabetic subjects.

The largest effect size was observed for TINN (Cohen's d = 1.05), followed by pNN50% (d = 0.98), indicating large magnitude differences between groups.

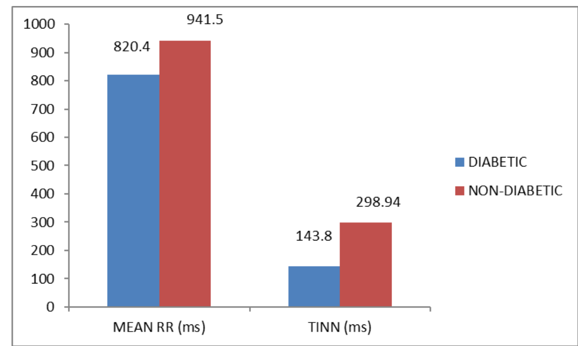

Figure 2: Comparison of Mean RR interval and TINN between cases and controls.

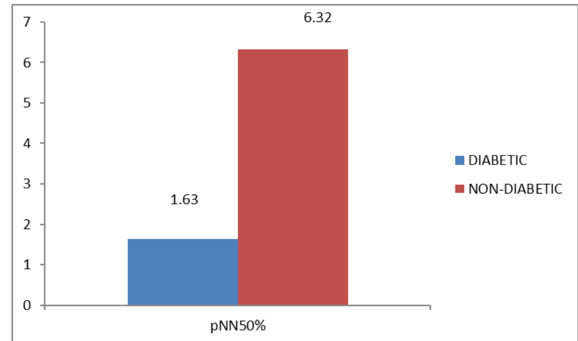

Figure 3: Comparison of pNN50% between cases and controls.

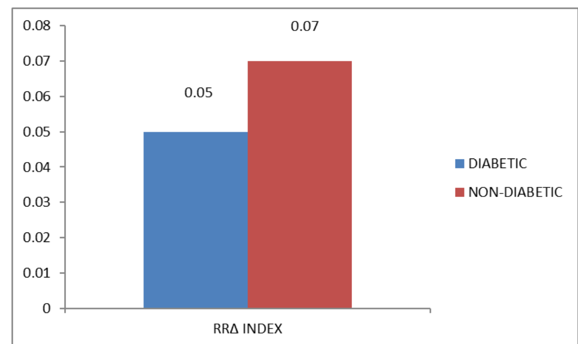

Figure 4: Comparison of RRΔ index between cases and controls.

5. Discussion

The present study demonstrated significant reduction in time-domain HRV parameters among asymptomatic T2DM patients, indicating early cardiac autonomic dysfunction. Reduced SDNN and RMSSD suggest parasympathetic withdrawal, while increased resting heart rate indicates relative sympathetic predominance [3,7]. These findings are consistent with recent meta-analyses demonstrating reduced HRV in T2DM [2,5]. Chronic hyperglycemia leads to oxidative stress and microvascular damage, contributing to autonomic nerve fiber injury [6]. Reduced HRV has also been associated with increased cardiovascular risk and mortality in diabetes [7,8]. A normal person exhibits a high

degree of beat-to-beat variability in heart rate, with HRV increasing during inspiration and decreasing during expiration. HRV reflects an individual's autonomic tone, where increased parasympathetic activity leads to bradycardia through enhanced vagal flow mediated by acetylcholine, thereby increasing beat-to-beat variation. Conversely, heightened sympathetic tone induces tachycardia by inhibiting vagal outflow from the cardiovascular center, reducing HRV [10-13]. Short-term HRV testing has shown reasonable diagnostic performance for early detection of CAN [2]. However, HRV should be interpreted alongside clinical evaluation [6]. Due to the cross-sectional design, causality cannot be established.

Limitations of the Study

- Moderate sample size
- Cross-sectional design
- HbA1c not recorded
- Frequency-domain HRV not evaluated

Future longitudinal studies are warranted.

6. Conclusion

Time-domain HRV parameters were significantly reduced in T2DM patients compared to healthy controls, reflecting early cardiac autonomic imbalance. Short-term HRV assessment may serve as a simple, non-invasive screening tool for early detection of subclinical CAN.

Ethical Clearance

This study was approved by the Institutional Ethics Committee of Yenepoya Medical College (YUEC498/2016). Written informed consent was obtained from all participants.

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Conflict of Interest

The authors declare no conflict of interest.

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