A COMPARATIVE STUDY OF HEART RATE VARIABILITY IN DIABETIC SUBJECTS AND NORMAL SUBJECTS

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Abstract

Aim: The main objective of this study is to enlighten medical faculty and diabetic patients about neurological and cardiovascular complications in Diabetes and to compare heart rate variability in normal individuals and diabetic patients.

Materials and Method: This research study was case-control study using Niviqure data acquisition system to record 5-minutes E.C.G. in 400 individuals who consisted of the cases group comprised of 200 diabetic patients and controls group consisted of 200 healthy individuals. 5-minutes E.C.G. data gathered was subjected to frequency domain analysis of Heart Rate Variability and from which various parameter depicting parasympathetic activity and sympathetic activity were analyzed. HbA1c levels were estimated by high performance liquid chromatography. Statistical analysis was done using MS office excel 2007 software.

Results: In the present study it was observed that parameters of heart rate variability are reduced (power of High Frequency-H.F., power of Low Frequency-L.F., total power-T.P. and L.F./H.F. ratio) and parameters of heart rate variability depicting parasympathetic modulation of heart (H.F.) are more reduced when compared to parameters of heart rate variability depicting sympathetic modulation (L.F.) in diabetics compared to normal individuals. It was also found that there is a negative correlation between HbA1c values of subjects and parameters of HRV.

Conclusion: These results may be attributed to early parasympathetic damage due to axonal degeneration of longer vagal fibers. This axonal degeneration is mostly caused due to chronically elevated levels of blood glucose.

Keywords: heart rate variability, diabetes mellitus, HbA1c, frequency domain analysis, FFT

1. Introduction

Current interest centers on the development of a new generation of tests of autonomic nerve function that are simple, non-invasive, reproducible and allow precision in diagnosis and accurate quantization. Most of them are based on cardiovascular reflexes and abnormality in them is assumed to reflect autonomic damage elsewhere. Measurement of Heart Rate Variability (HRV) is the best non-invasive method to measure the working of the heart, as it measures many aspects of cardiac functioning, including autonomic nerve functioning. The new method can replace the traditional manual method for evaluating cardiovascular responses with the advantages of speed and objectivity. Analysis of 5 minutes measurements of heart rate variability (HRV) has been shown to be a good predictor of physiological distress and mortality, especially for cardiovascular disease.

Diabetes is an ICEBERG disease. According to WHO report the prevalence of diabetes in adults worldwide has risen and the number will rise from 135 million in 1995 to 300 million by the year 2025. Epidemiological data in India shows the same upward trend according to the World Health Organization estimates, that India had 32 million diabetic subjects in the year 2000 and this number would increase to 80 million by the year 2030. Neuropathy is one of the most common complications of diabetes. Silent myocardial infarct is more common in diabetics due to involvement of cardiac autonomic nerves. At an early stage autonomic dysfunction may be asymptomatic or mildly symptomatic. Symptomatic autonomic neuropathy carries worst prognosis, so early diagnosis is essential for maximum benefit more so in diabetes. Heart rate variability monitoring plays a vital role in prevention and early diagnosis of cardiac autonomic neuropathic complications.
2. Material and method
Research has demonstrated that increased risk of cardiovascular disease (CVD) and sudden death due to M.I. exists in diabetic individuals. Because analysis of HRV has been used to predict cardiac morbidity and mortality, consequently this study was designed to investigate cardiovascular health of diabetic individuals using HRV.

2.1 Research design: This research study was case-control type of study where individuals suffering from diabetes mellitus were considered as cases and normal healthy individuals were considered as controls.

2.2 Size of sample: 400 individuals were selected for this study; both the study groups (case and controls) consisted of 200 subjects each. (20% of outpatient population for a period of six months)

2.3 Site of selection of sample: This study was carried out in the Departments of Physiology and Medicine at government hospital, Kakatiya Medical College, Warangal, Andhra Pradesh, India. Patients participating in the study had been referred to the Departments of Physiology from this hospital after determining their diabetic status, inclusion and exclusion criteria.

2.4 Determination of diabetic status: The criteria for diagnosis of diabetes mellitus are as follows. Symptoms of diabetes plus random blood glucose concentration $\geq 11.1$ mmol/L (200mg/dl) (OR) Fasting plasma Glucose $\geq 7.0$mmol/L (126mg/dl) (OR) Two-hour plasma glucose $\geq 11.1$ mmol/L (200 mg/dl) during an oral glucose tolerance test.

The same procedure was followed in control subjects to exclude the inclusion of asymptomatic diabetic subjects in control group.

2.5 Inclusion criteria:
1. Only normal healthy subjects, without any family history of diabetes mellitus, known chronic disease and not using any medicine for any reason, were included in the study as control group.
2. Established diabetic patients of both type I and type II were included in case group.
3. Confirmed diabetic patients whose blood sugar level was controlled on taking oral hypo-glycaemics were also included in the case group.

2.6 Exclusion Criteria:
1. History of Hypertension (sitting blood pressure $> 140/90$ mmHg).
2. History of alcohol / smoking.
3. History of intercurrent illness (e.g.- Pyrexia, Diarrhea).
4. History of drug intake.

5. Ages below 17 years and above 70 years.

Informed consent was taken from both groups by explaining what this study is meant for and ethical committee clearance was taken.

The controls and cases were clinically examined to rule out cardiovascular, respiratory, neurological and endocrinal disorders. Along with routine general examination, blood pressure height, weight, waist length recordings were taken. Blood samples were collected from subjects and controls. Hematological parameters were estimated in physiology department. The basal recording of blood pressure was done using sphygmomanometer by standard Riva Rocci method.

2.7 Recording of heart rate variability: The subjects were instructed to abstain from smoking and caffeine for 2 hrs. and alcohol for 36 hr. prior to the experiment, have adequate rest, get at least 8 hours of uninterrupted sleep on the night prior to the experiment, have a normal breakfast on the morning of the experiment and to void urine prior to the recording. All recordings were conducted between 10:30 A.M. to 1:00 P.M.

After comfortable strapping and rest for 5 min on the couch, ECG was recorded using disposable Ag/AgCl electrodes in standard lead II configuration.

A continuous recording of ECG signal was done for 5 min. From this recording suitable sections were taken out leaving behind the time of onset It was ensured that the temperature of the human physiology lab was comfortable before and after the E.C.G. recording. Heart rate variation during normal breathing for a period of 5 minutes was recorded, in supine position, awake and resting. ECG data in standard lead II configuration was acquired using portable ECG acquisition equipment (Niviqure Meditech Systems, Bangalore, India).

The data gathered was subjected to frequency domain analysis of heart rate variability HRV. Frequency domain analysis was performed using non-parametric method of Fast Fourier Transformation. Data was edited manually for artifacts and ectopic beats. HRV software (NIVIQURE ECG SOFTWARE for HRV studies Ver. 52.0.0) used a peak detection algorithm to find the ‘R’ wave, which was done at a resampling rate of ‘4 Hz’. A minimum of 256 data points was required to perform a spectral analysis. To attain 256 data points a duration of 5 minutes of ECG recording was required. The linear trend was removed from each data set to avoid its contribution to low
frequency power. The power frequency spectrum was subsequently quantified into 1.)Standard frequency – domain measurements as defined previously included total power (T.P.), HF (0.15–0.4 Hz), LF (0.04–0.15 Hz) and LF/HF ratio.

2.8 Biochemical test: Estimation of HBA1C was done by high performance liquid chromatography.HBA1C values are expressed as %.Normal value of HBA1C - <7% is considered normal.

Blood glucose (fasting blood sugar-FBS & post lunch blood sugar-PLBS) was determined on the auto analyzer by the Hexokinase method.Normal value of FBS-70-130 mg/dl; Normal value of PLBS<180 mg/dl.

2.9 Statistical Methods: Statistical analysis was performed with a Microsoft excel 2007 software. Data are expressed as mean ± SD. The procedure of testing the hypothesis was concluded by accepting the hypothesis or rejecting it. Data between the study group and control group were compared using z-test. When the statistic z value is computed, we found p value corresponding to it from the tables

If p value is < 0.001 it is considered highly significant and if it is <0.05 it is considered significant, and it is >0.05, then we concluded that the parameters under study are not significantly related.

In case of correlation study, Pearson’s correlation coefficient was used. Correlation value (r) lies in between -1 and +1. If the r value is -1≤ r <0-both parameters are negatively correlated If the r value is 0 < r ≤ 1 – both parameters are positively correlated if the r value is 0, no relation exists between the parameters.

3. Results

Table 1. Comparison of heart rate variability parameters in diabetics and normal individuals

| Parameters | Controls | Cases |
|------------|----------|-------|
| Mean | S.D.± | Mean | S.D.± |
| Age | 52.33 | 4.17 | 56.2 | 3.78 | 3.255 | 0.0014 |
| HbA1C | 6.1 | 0.44 | 8.29 | 0.90 | 12.13 | ≤ 0.0001 |
| F.B.S. | 85.1 | 7.38 | 128.56 | 15.54 | 13.88 | 0.0001 |
| P.L.B.S. | 131.5 | 9.46 | 201.06 | 35.69 | 10.27 | 0.0001 |
| Pulse rate | 77.03 | 6.65 | 85.33 | 8.15 | 4.045 | ≤ 0.00017 |
| S.B.P. | 119.74 | 8.52 | 136.66 | 8.61 | 8.298 | 1.89×10⁻⁹ |
| D.B.P. | 78.32 | 4.84 | 82.73 | 3.21 | 4.407 | 6.55×10⁻⁵ |
| L.F. | 327.63 | 165.35 | 189.1 | 114.93 | 3.803 | ≤ 0.00017 |
| H.F. | 187.4 | 79.93 | 97.2 | 71.56 | 4.573 | ≤ 0.00001 |
| L.F./H.F. | 1.83 | 0.64 | 2.13 | 0.54 | 1.72 | 0.04 |
| T.P. | 2011.86 | 2107.24 | 813.3 | 1456.49 | 2.560 | ≤ 0.006 |

Table 2. Correlation of heart rate variability parameters with HbA1C in both diabetics and normal individuals

| Parameter | L.F. | H.F. | L.F./H.F. | T.P. |
|-----------|------|------|-----------|------|
| HbA1C r-VALUE | -0.38 | -0.44 | 0.3 | -0.26 |

4. Discussion

In the present study it was observed that parameters of heart rate variability are reduced and parameters of heart rate variability depicting parasympathetic modulation of heart are more reduced when compared to parameters of heart rate variability depicting sympathetic modulation in diabetics compared to normal individuals. High frequency (H.F.) power parameter of frequency domain result more so in particular.

These parameter were chosen as a method for characterizing HRV in accordance with the recommendations of Task Force of the European Society of Cardiology and work done by Kitney RI et al, Akselrod et al, Bruce et al, Cowan M.J., Acharya R.U. et al, Pagani M. et al. These recommendations suggest that these parameters are sensitive measure of HRV and that these measures would primarily reflect parasympathetic mediated activity.

4.1. Parameters depicting parasympathetic activity: In our study power of High frequency (H.F.) values of frequency domain analysis results of H.R.V. are significantly reduced which is consistent with study done by H KUDAT, et al studied 31 diabetic patients and 30 age and sex matched controls, they found...
that parameters related to parasympathetic modulation of heart (H.F.) were significantly reduced.

4.2. Parameters of heart rate variability depicting sympathetic activity: Low frequency power measure of frequency domain result is a major quantitative marker of sympathetic activity \(^{19}\) was also significantly reduced in our study which is consistent with the study done by Pagani M, et al\(^ {20}\), H KUDAT et al\(^ {17}\) and Spallone V, et al\(^ {22}\). Studied power of low frequency parameter of frequency domain results for assessing sympathetic modulation of heart along with other parameters of heart rate variability in 35 normotensive diabetic patients. They found out that L.F. power values are significantly reduced in diabetic patient when compared to normal individuals, this consistent with our study.

4.3. Parameters of heart rate variability depicting total heart rate variability: Our study revealed that there significant reduction total heart rate variability indicated by Total power (T.P.) parameter of frequency domain analysis results which is consistent with the study done by Pagani M, et al\(^ {23}\) and H Kudat, et al\(^ {17}\).

4.4. Parameters of heart rate variability depicting sympatho-vagal balance: L.F./H.F. ratio which is marker of sympatho-vagal balance \(^ {23}\) was found significantly raised in diabetic patients when compared to normal individuals by Chessa M, et al\(^ {9}\). This is consistent with the results of L.F./H.F. ratio results found in our study. This means that power of HF is more reduced when compared to power of L.F. in diabetic individuals.

In this study both fasting blood sugar levels and post lunch blood sugar levels are raised in diabetes patients when compared to normal non-diabetic individuals.\(^ {6}\) This is in accordance with the recommendations of American diabetic association.\(^ {3}\)

4.5. Correlation of HbA1C with frequency domain results of H.R.V. in diabetes patients: The most significant factor correlating with incidence and progression of diabetic neuropathy is the status of Glycemic control (HBA1C). HBA1C reflects the status of glycaemia. In diabetes, presence of hyperglycaemia is the basis for pathogenesis and progression of autonomic neuropathy. In this study HbA1C levels in diabetic individuals are significantly raised. Colhoun, et al\(^ {11}\) in their study they concluded that reduced heart rate variability parameters may be a very early feature of insulin resistance and loss of glycemic control.

In this study HbA1c is negatively correlated (but not significant) with all parameters of heart rate variability (L.F., H.F. and T.P.) except L.F./H.F. ratio. It is positively correlated to L.F./H.F. ratio. This is consistent with study done by Faulkner MS, et al\(^ {14}\).

Conclusion

Our study suggests that there is both sympathetic dysfunction and parasympathetic dysfunction, but from increased values of L.F./H.F. ratios it is inferred that there is more parasympathetic damage than sympathetic nerve damage. Parasympathetic modulation is mediated via vagus nerve which itself is long nerve and it is well known fact that long nerves are prone for neuropathy in diabetes mellitus.\(^ {5}\) This may be accounted for early parasympathetic damage due to axonal degeneration of longer vagal fibers. This axonal degeneration is mostly caused due to chronically elevated levels of blood glucose. Reduced parasympathetic activity lowers the threshold for ventricular fibrillation, which is one of the important causes for sudden death.

Hence, the best way to prevent sudden death caused by cardiac autonomic neuropathy is by:-
1. Regularly monitoring HRV.
2. Maintaining good glycemic control.
3. Doing regular exercise increases parasympathetic tone.
4. Practicing yoga improves parasympathetic tone which reduces morbidity and mortality caused due cardiac autonomic neuropathy.

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