Autonomic neuropathy and plasma catecholamine in patients with diabetes mellitus

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Abstract

**Background:** cardiovascular autonomic neuropathy (CAN) is a common form of autonomic dysfunction in diabetes mellitus (DM) patients, but it can be asymptomatic for years. Low baseline plasma noradrenaline levels have been found in diabetic patients, but this decrease seems to associate clinically severe autonomic neuropathy.

**Purpose:** evaluate the impact of DM on heart rate variability (HRV) parameters and to determine the correlations with plasma adrenaline and noradrenaline, as a possible mechanism of early disruption in HRV.

**Methods:** a group of 34 patients with type 2 diabetes mellitus, without clinical signs of CAN, was enrolled. HRV (as a measure of autonomic balance) was measured using a 24-hour ECG monitoring system in all subjects during normal daily activity. Plasma catecholamines and other laboratory markers were measured.

**Results:** HRV parameters are lower in DM group as compared with control group. More than half of the patients had HRV parameters below the normal range (54%). There are lower levels of noradrenaline value in DM, as compared with controls, but the difference is not statistically significant (p = 0.08). Adrenaline levels were similar in both groups.

**Discussions and conclusion:** CAN is best evaluated using heart rate variability (HRV) on 24-hours recordings. There is a tendency for HRV parameters to decrease even in asymptomatic patients, especially after years of evolution. We didn’t find significant correlations between HRV and plasma catecholamine, even if noradrenaline was lower in DM patients. Holter monitoring remains a reliable method for early diagnosis of CAN.

**Keywords:** diabetes mellitus, heart rate variability, plasma catecholamine
Introduction

The autonomic nervous system is often imbalanced in patients with type 2 diabetes mellitus (DM) and this may be clinically inapparent. Cardiovascular autonomic neuropathy (CAN) is a common form of autonomic dysfunction in DM patients and associates abnormalities in heart rate control, as decreased heart rate variability (HRV) and in central and peripheral vascular dynamics [1].

Plasma catecholamines generally provide an index of sympathetic neural activity (noradrenaline), or an index of sympatho-adrenal activity (adrenaline) [2]. Under normal conditions, these hormones show a circadian rhythm with higher levels during daytime activity and lower levels during the night [3, 4]. Subnormal baseline plasma noradrenaline levels have been found in diabetic patients [5], but this decrease is only found in patients with a long duration of diabetes, with clinically severe autonomic neuropathy. On the other hand, is well known the fact that cardiovascular system, including HRV, is under sympatho-vagal control, so plasma catecholamines level could be an indicator of altered HRV in diabetics [5, 6].

At the time of diagnosis, a reduced HRV is evident in type 2 DM which reflects the asymptomatic process over many years, before diagnosis [6, 7, 8]. Autonomic nervous function can be assessed according to the consensus statement of the American Diabetes Association and the American Academy of Neurology, using four tests, the description of which is beyond the interest of this study; 24-hour ECG recording is another method used more and more in the last decade, as an alternative of above mentioned tests, and seems more reliable and more sensitive [9, 10, 11].

The objective of this study was to evaluate the impact of diabetes mellitus on heart rate variability parameters measured on 24-hour ECG recording in a group of type 2 diabetes mellitus, and to determine the correlations, if any, with plasma adrenaline and noradrenaline, as a possible mechanism of early disruption in heart rate variability.
Subjects and methods

Study population

The study group consisted of 34 patients, males and females, diagnosed with type 2 diabetes mellitus and followed up at an outpatient clinic. There were excluded from the study patients with chronic heart failure, chronic kidney failure, hypertension, and body mass index over 25 kg/m². The patients included in the study had no clinical signs of autonomic neuropathy. The control group consisted of 29 healthy subjects who were matched for age and sex.

Informed consent was obtained from all patients and the study was conducted according to the Declaration of Helsinki.

Heart rate variability

HRV was measured using a 24-hour ECG monitoring system (Holter Digital recorder AsPEKT 812) in all subjects during normal daily activity. Time domain parameters used are: SDNN expressed in milliseconds (ms) accounts for standard deviation of all NN intervals. SDANN expressed in ms accounts for standard deviation of the averages of NN intervals in all 5 min segments of the entire recording. pNN50 % is the number of pairs of adjacent NN intervals divided by the total number of all NN intervals. Frequency domain parameters used are: low frequency and high frequency components of spectral analysis expressed in squared milliseconds (ms² or normalized units [11].

Laboratory markers

Blood was collected into chilled tubes containing sodium ethylene-diamine tetracetic acid for measurement of the plasma adrenaline and noradrenaline levels. Plasma was separated within 10 min in a refrigerated centrifuge. Samples were then immediately placed in a freeze and stored at −70°C until assayed. Plasma adrenaline and noradrenaline levels were measured by high performance liquid chromatography [12]. Samples were collected at 6.00, 18.00, and 03.00 o’clock. There were also measured: fasting plasma glucose, cholesterol, triglycerides and glycated hemoglobin (HbA1), from the morning blood sample, collected in separate tubes.
**Statistical analysis**

The data are presented as mean ± SD unless otherwise specified. Comparison between groups of subjects for various parameters was performed by ANOVA using SPSS 8 for Windows. Student’s paired t test and Pearson’s linear correlation coefficients were also used to evaluate the data (for statistical significance and for pairs of continuous variables). A p value less than 0.05 was considered statistically significant.

**Results**

Characteristics of the DM patients are shown in table 1.

**Place for table 1**

*Heart rate variability parameters*

Heart rate variability parameters are lower in DM group as compared with control group, but differences are significant only for SDNN (from time domain parameters) and LF, HF, and normalized units HF (for frequency domain parameters). More than half of the patients had HRV parameters below the normal range (54%).

**Place for table 2**

*Laboratory measurements*

A circadian variation with higher values in the daytime was evident in both groups. There are no differences between day/night values of adrenaline or noradrenaline in the two groups. Also, adrenaline values were similar in both groups. Diabetic patients exhibited a tendency for a lower noradrenaline value than did the controls, but the difference is not statistically significant (p = 0.08).

*Correlations*

There was a mild positive correlation between the mean 24-h plasma noradrenaline level and heart rate (r = 0.35, p = 0.040). There were no significant correlations between plasma noradrenaline and LF/HF (which accounts for sympatho-vagal balance), LF (which accounts for both sympathetic and parasympathetic activity) or HF (which accounts for vagal activity).

**Discussions**

CAN is best evaluated using heart rate variability (HRV) on 24-hours recordings. A reduction in time-domain parameters of heart rate variability seems not only to carry negative prognostic value but also to precede the clinical expression of autonomic...
neuropathy [11, 13, 14, 15]. In diabetic patients without evidence of autonomic neuropathy, reduction of the absolute power of low-frequency (LF) and high-frequency (HF) during controlled conditions was reported [11]. LF component accounts for both sympathetic and vagal dysfunction, very-low frequency (VLF) component accounts for sympathetic dysfunction, and HF component accounts for parasympathetic dysfunction. However, when the LF/HF ratio is considered or when LF and HF are analyzed in normalized units, no significant difference in comparison to normal individuals is present [11]. In advanced cardiac autonomic neuropathy all the components of HRV are reduced (both for sympathetic and parasympathetic activity), along with LF/HF ratio [11, 16].

In our study, we found a positive correlation between disease duration and HRV parameters. The strongest correlation was between HF and disease duration (r = 0.75). The correlation between disease duration and autonomic damage is still under debate [10]. As we already said, at the time of diagnosis, a reduced HRV is frequently discovered in type 2 DM patients, which reflects the manifestation of the asymptomatic process during many years [10, 11, 17]. The connection between autonomic neuropathy, disease duration of diabetes and patient’s age are still unclear [6]. On the other hand, even the symptoms and signs of autonomic dysautonomy may be stable over time, both in type 1 and type 2 DM patients [6]. This suggests the need of reliable, objective diagnostic tools, other than clinical ones, to allow early risk stratification [10, 11, 16].

The present study showed that diabetic patients have also lower, even if non-significant, levels of plasma noradrenaline compared with controls. Catecholamines are neurotransmitters whose circulating levels reflect sympathetic nervous system activity. Noradrenaline is mainly released from the sympathetic nerve endings in response to nerve impulses, while adrenaline is secreted from the adrenal medulla upon sympathetic activation [18]. There are still some controversies concerning the plasma catecholamine levels in diabetic patients. Some investigators have described normal or elevated plasma catecholamines [19, 20], and others, like us, have found a reduction in the circulating concentrations of noradrenaline, in different extents [21, 22, 23]. There are studies showing a depletion of noradrenaline stores in the cardiovascular system [24, 25] and destruction of distal postganglionic sympathetic nerves in diabetic patients [26, 27]. To summarize, in our study, plasma catecholamines, even if noradrenaline showed lower
values (p = 0.08), had no significant correlations with HRV parameters; it seems that in early stages of CAN, 24 hours Holter ECG registration remains the most valuable and reliable method, being also not expensive and non invasive.

A limitation of this study may be the dimension of the study group; it is possible that in a much larger group the results were different regarding plasma noradrenaline levels and/or correlations between catecholamines and HRV parameters. Another limitation could be hormone measurement; there are studies providing a 4-hour interval blood prelevation [24, 27], which may be more accurate for a 24 h evaluation of plasma catecholamines.

**Conclusion**

It is important to remember that autonomic dysfunction can be detected at the time of diagnosis of DM, and correlates with poor glycemic control; there is a long period of time in which patients are asymptomatic but the disease progress; this so called subclinical autonomic neuropathy should be checked using autonomic function tests, including 24-hour Holter monitoring. Waiting for symptoms to appear is not recommended, as clinical signs and symptoms are not reliable tools in early diagnosis risk stratification. Also, early alterations in plasma levels of noradrenaline may explain the sympathto-vagal imbalance in these patients.
References


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