Heart Rate Variability and Heart Rate Turbulence in HIV-Infected Patients Receiving Combination Antiretroviral Therapy

Wanwarang Wongcharoen, M.D.,*‡ Kolakrit Khienprasit, M.D.,†
Arintaya Phrommintikul, M.D.,*‡ Apichard Sukonthasarn, M.D.,*
and Nipon Chattipakorn, M.D., Ph.D.‡

From the *Department of Internal Medicine, Faculty of Medicine, Chiang Mai University, Chiang Mai, Thailand; †McCormick Hospital, Chiang Mai, Thailand; and ‡Cardiac Electrophysiology Research and Training Center, Department of Physiology, Faculty of Medicine, Chiang Mai University, Chiang Mai, Thailand

Background: Previous studies have demonstrated the presence of autonomic dysfunction in human immunodeficiency virus (HIV)-infected patients. However, the data in those receiving combination antiretroviral therapy (cART) are conflicting. The aim of this study was to assess the autonomic function using heart rate variability (HRV) and heart rate turbulence (HRT) analysis in HIV-infected patients receiving cART.

Methods: Eighty-one HIV-infected patients receiving cART and 42 control subjects were enrolled in the study. The HRV and HRT parameters were assessed on 24-hour digital Holter electrocardiogram recordings.

Results: Baseline characteristics were comparable between HIV-infected and control subjects, except the higher fasting glucose and triglyceride and lower high-density lipoprotein cholesterol observed in HIV-infected patients. All components of HRV were significantly reduced in HIV-infected patients. After adjustment with biochemical parameters, most of the HRV parameters were still significantly reduced in HIV-infected patients. However, HRV parameters reflecting vagal activity were no longer different between 2 groups. In addition, HRT parameters did not differ between HIV-infected and control subjects. The standard deviation of normal-to-normal intervals significantly correlated with CD4 lymphocyte counts in HIV-infected patients but did not with protease inhibitors therapy.

Conclusions: We demonstrated the overall decrease in HRV in HIV-infected patients receiving cART. The metabolic disturbance observed in HIV-infected patients possibly accounted for decreased vagal activity.

Ann Noninvasive Electrocardiol 2013;18(5):450–456

antiretroviral therapy; autonomic dysfunction; human immunodeficiency virus; heart rate turbulence; heart rate variability

INTRODUCTION

Human immunodeficiency virus (HIV) infection is a significant health problem worldwide. Even though the mortality rate from acquired immune

deficiency syndrome (AIDS) has decreased dramatically after the development of combination antiretroviral therapy (cART), the cardiovascular death has been found increased in HIV-infected patients.^{1–5} Sudden cardiac arrest contributes to

Address for correspondence: Wanwarang Wongcharoen, M.D., Department of Internal Medicine, Faculty of Medicine, Chiang Mai University, Chiang Mai, Thailand. Fax: +66-53-945486; E-mail: bwanwarang@yahoo.com

This work was supported by grants from the National Research University Project under Thailand's Office of the Higher Education Commission, Thailand Research Fund MRG 5380258 (W.W.), MRG 5280169 (A.P.), RTA 5280006 (N.C.), and the Faculty of Medicine Endowment Fund for medical research, Chiang Mai University, Thailand (W.W., A.P., and N.C.).

Conflicts of interest: The authors report no conflicts of interest.

most cardiac deaths in HIV-infected patients with a rate more than four times expected in the general population with similar risk factors.^{6,7} It has been suggested that autonomic dysfunction may be the mechanism underlying sudden cardiac death in this group of patients.^{8–11} Previous small studies have demonstrated the presence of autonomic dysfunction in untreated HIV-infected patients.¹² Since the introduction of effective treatment regimes, the viral suppression and metabolic disturbances induced by cART may have an additional effect on the autonomic function. Few studies have examined the autonomic function using short-term heart rate variability (HRV) in HIV-infected patients on cART and the results are conflicting.^{13–15} The 24-hour HRV has been shown to be more reliable than the short-term HRV.^{16,17} In addition, the heart rate turbulence (HRT), which reflects baroreflex activity has never been studied in HIV-infected patients previously.¹⁸ Therefore, we conducted the study to assess the 24-hour HRV and HRT in HIV-infected patients receiving cART. Other potential factors, which may have affected the autonomic function, were also examined.

METHODS

Studied Population

Between February 2009 to January 2012, 81 HIV-infected patients (age >18 years) attending the HIV clinic at Maharaj Nakorn Chiang Mai Hospital were enrolled. All HIV-infected patients had been receiving combination of at least three antiretroviral agents selected from several drug classes, which included nucleoside reverse transcriptase inhibitors (NRTIs), non-NRTIs (NNRTIs), and protease inhibitors (PIs). Patients were excluded if they had apparent heart diseases, diabetes mellitus, atrial flutter, atrial fibrillation, thyrotoxicosis, pacemaker installation, or had been taking any medications, which affected autonomic function including contraceptive, tricyclic antidepressant, anticholinergic agents, amphotericin B, calcium channel blocker, beta-blocker, antiarrhythmic, and centrally acting antihypertensive agents. The CD4 lymphocyte counts and HIV RNA levels were carried out in all HIV-infected patients. Forty-two healthy volunteers with a negative HIV antibody test were also recruited in the study. History taking, physical examination, 12-lead electrocardiography (HP M1700A, Hewlett Packard, Palo

Alto, CA, USA), and echocardiography (Philips iE33, Philips Ultrasound, Bothell, WA, USA) were performed to exclude structural heart diseases. All subjects underwent 24-hour Holter (GE Seer Light Extend, GE Medical Systems, Suzuken Company, Limited, Nagoya, Aichi, Japan) monitoring for HRV and HRT assessment. Recordings lasting for ≥ 16 hours and of sufficient quality for evaluation were included in the analysis. Before analyzing the data, they were manually preprocessed. The study protocol was approved by the local institutional review board. Informed consent was obtained from all subjects.

HRV

Time-domain and frequency-domain analyses were performed according to the standard guidelines.¹⁹ Time-domain HRV indexes were analyzed using statistical methods. The square root of the mean squared differences of successive normal-to-normal (NN) intervals (rMSSD), the standard deviation (SD) of all NN intervals (SDNN), the average of the SD of the 5-minute NN intervals over the entire recording (ASDNN), the SD of the average NN intervals calculated over 5-minute periods of the entire recording (SDANN), and the proportion of adjacent NN intervals differing by >50 ms (pNN50) in the 24-hour recording were measured.

Frequency-domain HRV were analyzed using autoregressive power spectral analysis applied to the RR interval time series. The following spectral bands were identified: very low frequency (VLF; 0.003–0.04 Hz), low frequency (LF; 0.04–0.15 Hz), and high frequency (HF; 0.15–0.4 Hz). Total power (0–0.5 Hz) and the areas below each peak were calculated in absolute units (ms^2).

HRT

Two parameters describing HRT were calculated.¹⁸ Turbulence onset (TO) was defined as the difference between the mean of the first two sinus RR intervals after ventricular premature contraction (VPC), and the last two sinus RR intervals before VPC divided by the mean value of the last two sinus RR intervals before VPC. TO (expressed in%) was calculated for each VPC and then averaged and a TO value <0% indicates early sinus acceleration and is considered normal. Turbulence slope (TS) was defined as the maximum positive value of the slope of a regression line assessed over any sequence of five subsequent sinus RR

Table 1. Baseline Characteristic of the Studied Subjects

Characteristics	HIV-Infected (n = 81)	Control (n = 42)	P Value
Age (years)	44 ± 8	43 ± 9	0.518
Male	36 (44%)	16 (38%)	0.566
FBG (mg/dL)	100.46 ± 23.83	88.50 ± 12.38	0.002
Total cholesterol (mg/dL)	212.14 ± 40.33	193.68 ± 40.85	0.067
Triglyceride (mg/dL)	178.88 ± 120.48	111.71 ± 70.60	<0.001
LDL cholesterol (mg/dL)	130.93 ± 44.81	117.96 ± 31.65	0.162
HDL cholesterol (mg/dL)	54.82 ± 26.84	58.21 ± 13.33	0.050
Creatinine (mg/dL)	0.94 ± 0.23	0.95 ± 0.20	0.667
LVEF (%)	66.24 ± 10.19	67.77 ± 10.00	0.767

FBG = fasting blood glucose; HDL = high-density lipoprotein; LDL = low-density lipoprotein; LVEF = left ventricular ejection fraction.

intervals within the first 20 sinus intervals after VPC. TS was calculated based on an averaged local tachogram and a TS value >2.5 ms/RR interval indicates the normal expected late deceleration. According to the standard measurement of HRT, we excluded the subjects who had <5 VPCs during 24-hour recording from the HRT analysis for the reliable construction of the VPC tachogram.

Statistical Analysis

Results were expressed as mean ± SD, unless otherwise specified. The numerical variables were compared between groups with the Mann-Whitney U test. Proportions were compared by Fisher's exact test. The comparisons of HRV and HRT parameters between groups were adjusted according to potential confounding variables. Multivariate analyses were performed for variables with a P value <0.1 in univariate analysis using the linear regression procedure. Correlations were analyzed using nonparametric (Spearman) correlation and expressed with Spearman's rho. P values <0.05 were considered statistically significant. Statistical software package SPSS version 15.0 (Chicago, IL, USA) was used for analysis.

RESULTS

Baseline Characteristics

The baseline characteristics were comparable between HIV-infected patients and control subjects in terms of age, gender, creatinine, left ventricular systolic function. However, the fasting blood glucose and triglyceride levels were significantly higher in HIV-infected patients than control

subjects. In addition, the high-density lipoprotein (HDL) cholesterol was lower in HIV-infected patients (Table 1). The other parameters of HIV-infected patients are shown in Table 2.

HRV Analysis

The results of HRV are demonstrated in Table 3. All parameters of time- and frequency-domain measurements except LF/HF ratio were significantly lower in HIV-infected patients when compared with the control group. After multivariate linear regression analysis adjusted with fasting blood glucose, HDL cholesterol, and triglyceride, HIV-infected patients still had significantly lower SDNN, SDANN, ASDNN, total power, VLF, and LF, compared to control subjects. In addition, the reduction in LF/HF ratio in HIV-infected patients became statistically significant after the multivariate analysis. However, rMSDD, pNN50, and HF were no longer different between HIV-infected

Table 2. HIV Parameters in HIV-Infected Group

HIV Parameters	N = 81
HIV duration (years)	7.4 (5.5–12.1)
cART duration (years)	3.8 (2.0–5.5)
CD4+ cell count (cells/mm ³)	415 (334–608)
HIV RNA <50 copies/mL	72 (88.9%)
HIV RNA <400 copies/mL	79 (97.5%)
HAART regimens	
NRTIs	3 (3.7%)
NRTIs + NNRTIs	68 (84.0%)
NRTIs + PI	10 (12.3%)

Values are median (interquartile range).

cART = combination antiretroviral therapy; HIV = human immunodeficiency virus; NRTI = nucleoside reverse-transcriptase inhibitor; NNRTI = nonnucleoside reverse-transcriptase inhibitor; PI = protease inhibitor.

Table 3. Univariate and Multivariate Analyses of Heart Rate Variability between HIV-Infected Patients and Controls

Parameters	HIV-Infected (N = 81)	Control (N = 42)	P Value	Adjusted P Value ^a
SDNN (ms)	109.94 ± 28.12	127.86 ± 30.16	0.002	0.026
SDANN (ms)	102.22 ± 29.39	120.98 ± 33.19	0.002	0.015
ASDNN (ms)	40.64 ± 11.49	49.93 ± 12.50	<0.001	0.014
rMSSD (ms)	23.96 ± 9.36	29.10 ± 9.60	0.004	0.318
pNN50 (%)	5.53 ± 6.09	8.94 ± 8.10	0.005	0.418
Total power (ms ²)	2870.93 ± 844.87	3589.76 ± 940.66	<0.001	0.011
VLF power (ms ²)	22.53 ± 6.23	27.45 ± 6.97	<0.001	0.013
LF power (ms ²)	14.10 ± 5.57	18.21 ± 5.85	<0.001	0.002
HF power (ms ²)	9.64 ± 4.39	12.21 ± 4.58	0.001	0.343
LF/HF ratio	1.51 ± 0.43	1.55 ± 0.37	0.312	0.015

^aMultivariate linear regression adjusted by fasting blood glucose, high-density lipoprotein cholesterol, and triglyceride. SDNN = standard deviation (SD) of all normal-to-normal (NN) intervals; SDANN = SD of the average NN intervals calculated over 5-minute periods of the entire recording; ASDNN = average of the SD of the 5-minute NN intervals over the entire recording; rMSSD = square root of the mean squared differences of successive NN intervals; pNN50 = proportion of adjacent NN intervals differing by >50 ms in the 24-hour recording; VLF = very low frequency; LF = low frequency; HF = high frequency.

patients and controls after the adjustment (Table 3). We found that triglyceride was significantly associated with the lower HF in the multivariate analysis ($P = 0.044$).

HRT Analysis

The HRT could be analyzed only in 30 of 81 (37%) HIV-infected patients and 19 of 42 (45%) control subjects due to the lack of VPCs in the rest of the subjects. TO did not differ between HIV-infected and control subjects ($-1.5 \pm 2.9\%$ vs $-2.4 \pm 2.8\%$, respectively, $P = 0.157$). Furthermore, there was no difference of TS between two groups (10.6 ± 12.5 vs 10.3 ± 5.2 ms/RR interval, $P = 0.119$).

The Correlation between the Autonomic Function and HIV Parameters

We examined the correlation between the autonomic function and HIV parameters including CD4+ cell count, HIV duration, and cART duration. We demonstrated that CD4+ cell count had a weakly positive correlation with SDNN ($r = 0.246$, $P = 0.027$; Fig. 1A) and with SDANN ($r = 0.250$, $P = 0.024$; Fig. 1B). However, there was no correlation between autonomic function and either HIV duration or cART duration.

The Correlation between the Autonomic Function and Biochemical Parameters

We also investigated the correlation between the autonomic function and biochemical parameters.

We found that triglyceride correlated inversely with ASDNN ($r = -0.208$, $P = 0.037$), with total power ($r = -0.221$, $P = 0.026$), with VLF ($r = -0.200$, $P = 0.045$), and with HF ($r = -0.229$, $P = 0.021$). There were no significant correlations between autonomic function and either fasting blood glucose or cholesterol levels.

The Comparison between HIV-Infected Patients with Protease-Inhibitor Therapy and Nonprotease-Inhibitor Therapy

Since the exposure to PI-based therapy has been associated with an increased risk of myocardial infarction and sudden death,^{20,21} we also examined whether there were any differences in biochemical parameters and autonomic function between HIV-infected patients who received PI-based regimens and those who received non-PI-based regimens. We found that the patients with PI-based therapy had nonsignificantly lower triglyceride (134 ± 88 mg/dL vs 185 ± 124 mg/dL, $P = 0.076$) and lower HDL cholesterol (44 ± 11 mg/dL vs 57 ± 28 mg/dL, $P = 0.060$). Nevertheless, there were no disparities in the autonomic function between the two groups.

DISCUSSION

The use of cART has markedly improved survival in HIV-infected patients.⁵ However, there is a growing recognition of increased cardiovascular risk among human HIV-infected patients.^{3,4} It is well established that autonomic dysfunction plays an important role in the occurrence of

Figure 1. The correlation between CD4 count and (A) standard deviation of normal-to-normal intervals (SDNN) and (B) standard deviation of the average NN intervals calculated over 5-minute periods of the entire recording (SDANN) in 81 HIV patients receiving highly active antiretroviral treatment.

cardiovascular morbidity and mortality.^{11,22} Few studies have assessed the autonomic function in cART-receiving HIV-infected patients. Correia et al. demonstrated that there were no autonomic alterations observed in 40 AIDS patients on cART when compared with the control group.¹³ Conversely, other two studies of cART-treated HIV-infected patients showed that autonomic dysfunction was present in those patients.^{14,15} However, all those studies used short-term HRV to assess autonomic function.^{13–15} It has been shown that 24-hour HRV is more reliable than the short-term HRV.¹⁶ In this study, we found that HIV-infected patients on cART had a reduction in all parameters of 24-hour HRV. We demonstrated that HIV-infected patients had higher fasting blood glucose and triglyceride and lower HDL cholesterol than the controls, similar to other previous studies.^{15,20} The metabolic disturbances observed in HIV-infected patients have been described to be associated with antiretroviral drug therapy.^{15,20}

After the adjustment with biochemical parameters, the SDNN, total power, and LF were still significantly reduced in the HIV-infected patients. In addition, the decrease in LF/HF ratio in HIV-infected patients became statistically significant after the adjustment. However, the rMSDD, pNN50, and HF were no longer different between HIV-infected and control groups after multivariate analysis. On the other hand, the higher triglyceride was found to be independently associated with the lower HF. Short-term HRV variables including

rMSDD, pNN50, and HF are considered to primarily reflect the short-term effects of vagal activity on cardiac autonomic function. Thus, the presence of metabolic disturbances may have partly accounted for the blunted vagal activity demonstrated in HIV-infected patients. This finding is in accordance with that of a previous study, which showed that the decreased parasympathetic activity has been associated with the metabolic abnormalities.²³

Similar to the short-term HRV variables, the HRT also represents short-term regulation of vagal activity.¹⁸ As we demonstrated that there were no differences of HRT parameters noted between the HIV-infected and control group, these findings supported that the HIV infection per se may not have had an effect on the short-term vagal activity. However, we could assess HRT in the limited number of the subjects due to the lack of VPCs, therefore, a study with larger population is needed to confirm this finding.

The pathophysiologic mechanisms of autonomic dysfunction in HIV-infected are not fully elucidated. There are several possible factors that have been proposed. The first explanation is the neurotropic effect of HIV virus itself.^{24,25} However, we demonstrated only weakly positive correlation between autonomic function and immune status, which indicated that this factor contributed only in a limited way in determining cardiac autonomic dysfunction. Second, the autonomic dysfunction may be due to the adverse effects of longstanding treatment of antiretroviral therapy related to the

neuropathy.^{26–28} Moreover, the effect of cART on metabolic disturbances could lead to parasympathetic damage as it is seen in early diabetes and metabolic syndrome.²³ However, we did not find any association between different classes of cART and autonomic function.

This study had some limitations. We did not have the HRV data of the HIV-infected patients before starting of cART. Therefore, the change of HRV before and after cART could not be evaluated. This information may help clarify whether the change of HRV is secondary to therapy or the HIV infection per se. In addition, due to the relatively young age of the study population, we demonstrated the low cardiovascular events in the HIV-infected patients. With this regard, the association between abnormal HIV parameters and clinical outcome could not be assessed. Long-term study is required to clarify this issue.

CONCLUSION

We demonstrated the presence of autonomic dysfunction in HIV-infected patients receiving cART. The overall decrease in HRV was observed in this group of patients. The metabolic disturbance observed in HIV-infected patients possibly accounted for the decreased vagal activity. A weak correlation between autonomic function and CD4+ cell count was also demonstrated. It is possible that the interventions, which improve immune status and metabolic disturbances, may help improve the autonomic function in HIV-infected patients receiving cART.

REFERENCES

1. Friis-Moller N, Sabin CA, Weber R, et al. Combination antiretroviral therapy and the risk of myocardial infarction. *N Engl J Med* 2003;349:1993–2003.
2. Hsue PY, Giri K, Erickson S, et al. Clinical features of acute coronary syndromes in patients with human immunodeficiency virus infection. *Circulation* 2004;109:316–319.
3. Aberg JA. Cardiovascular complications in HIV management: Past, present, and future. *J Acquir Immune Defic Syndr* 2009;50:54–64.
4. Currier JS. Update on cardiovascular complications in HIV infection. *Top HIV Med* 2009;17:98–103.
5. Lewden C, May T, Rosenthal E, et al. Changes in causes of death among adults infected by HIV between 2000 and 2005: The "Mortalite 2000 and 2005" surveys (ANRS EN19 and Mortavic). *J Acquir Immune Defic Syndr* 2008;48:590–598.
6. Tseng ZH, Secemsky EA, Dowdy D, et al. Sudden cardiac death in patients with human immunodeficiency virus infection. *J Am Coll Cardiol* 2012;59:1891–1896.
7. Chugh SS, Jui J, Gunson K, et al. Current burden of sudden cardiac death: Multiple source surveillance versus retrospective death certificate-based review in a large U.S. community. *J Am Coll Cardiol* 2004;44:1268–1275.
8. Welby SB, Rogerson SJ, Beeching NJ. Autonomic neuropathy is common in human immunodeficiency virus infection. *J Infect* 1991;23:123–128.
9. Villa A, Foresti V, Confalonieri F. Autonomic neuropathy and prolongation of QT interval in human immunodeficiency virus infection. *Clin Auton Res* 1995;5:48–52.
10. Puri R, Roberts-Thomson KC, Young GD. HIV and long QT syndrome—cause or coincidence. *Int J Cardiol* 2009;133:e9–e10.
11. Pokorny J, Stanek V, Vrana M. Sudden cardiac death thirty years ago and at present. The role of autonomic disturbances in acute myocardial infarction revisited. *Physiol Res* 2011;60:715–728.
12. Mittal CM, Wig N, Mishra S, et al. Heart rate variability in human immunodeficiency virus-positive individuals. *Int J Cardiol* 2004;94:1–6.
13. Correia D, Rodrigues De Resende LA, Molina RJ, et al. Power spectral analysis of heart rate variability in HIV-infected and AIDS patients. *Pacing Clin Electrophysiol* 2006;29:53–58.
14. Lebech AM, Kristoffersen US, Mehlsen J, et al. Autonomic dysfunction in HIV patients on antiretroviral therapy: Studies of heart rate variability. *Clin Physiol Funct Imaging* 2007;27:363–367.
15. Askgaard G, Kristoffersen US, Mehlsen J, et al. Decreased heart rate variability in HIV positive patients receiving antiretroviral therapy: Importance of blood glucose and cholesterol. *PLoS One* 2011;6(5):e20196.
16. Assoumou HG, Pichot V, Barthelemy JC, et al. Metabolic syndrome and short-term and long-term heart rate variability in elderly free of clinical cardiovascular disease: The PROOF study. *Rejuvenation Res* 2010;13:653–663.
17. Chattipakorn N, Incharoen T, Kanlop N, et al. Heart rate variability in myocardial infarction and heart failure. *Int J Cardiol* 2007;120:289–296.
18. Bauer A, Malik M, Schmidt G, et al. Heart rate turbulence: Standards of measurement, physiological interpretation, and clinical use: International Society for Holter and Noninvasive Electrophysiology Consensus. *J Am Coll Cardiol* 2008;52:1353–1365.
19. Camm AJ, Malik M, Bigger JT, et al. 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. *Eur Heart J* 1996;17:354–381.
20. Friis-Moller N, Reiss P, Sabin CA, et al. Class of antiretroviral drugs and the risk of myocardial infarction. *N Engl J Med* 2007;356:1723–1735.
21. Worm SW, Kamara DA, Reiss P, et al. Evaluation of HIV protease inhibitor use and the risk of sudden death or nonhemorrhagic stroke. *J Infect Dis* 2012;205:535–539.
22. Becker K, Grolach I, Frieling T, et al. Characterization and natural course of cardiac autonomic nervous dysfunction in HIV-infected patients. *AIDS* 1997;11:751–757.
23. Licht CM, Vreeburg SA, van Reedt Dortland AK, et al. Increased sympathetic and decreased parasympathetic activity rather than changes in hypothalamic-pituitary-adrenal axis activity is associated with metabolic abnormalities. *J Clin Endocrinol Metab* 2010;95:2458–2466.
24. Ruttimann S, Hilti P, Spinass GA, et al. High frequency of human immunodeficiency virus-associated autonomic

- neuropathy and more severe involvement in advanced stages of human immunodeficiency virus disease. *Arch Intern Med* 1991;151:2441–2443.
25. Melli G, Keswani SC, Fischer A, et al. Spatially distinct and functionally independent mechanisms of axonal degeneration in a model of HIV-associated sensory neuropathy. *Brain* 2006;129:1330–1338.
26. Dalakas MC. Peripheral neuropathy and antiretroviral drugs. *J Peripher Nerv Syst* 2001;6:14–20.
27. Peltier AC, Russell JW. Recent advances in drug-induced neuropathies. *Curr Opin Neurol* 2002;15:633–638.
28. Authier FJ, Gheradi RK. Peripheral neuropathies in HIV-infected patients in the era of HAART. *Brain Pathol* 2003;13:223–228.