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ORIGINAL RESEARCH

Heart Rate Variability, Deceleration Capacity of Heart Rate, and Death: A Veteran Twins Study

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BACKGROUND: Autonomic function can be measured noninvasively using heart rate variability (HRV), which indexes overall sympathovagal balance. Deceleration capacity (DC) of heart rate is a more specific metric of vagal modulation. Higher values of these measures have been associated with reduced mortality risk primarily in patients with cardiovascular disease, but their significance in community samples is less clear.

METHODS AND RESULTS: This prospective twin study followed 501 members from the VET (Vietnam Era Twin) registry. At baseline, frequency domain HRV and DC were measured from 24-hour Holter ECGs. During an average 12-year follow-up, all-cause death was assessed via the National Death Index. Multivariable Cox frailty models with random effect for twin pair were used to examine the hazard ratios of death per 1-SD increase in log-transformed autonomic metrics. Both in the overall sample and comparing twins within pairs, higher values of low-frequency HRV and DC were significantly associated with lower hazards of all-cause death. In within-pair analysis, after adjusting for baseline factors, there was a 22% and 27% lower hazard of death per 1-SD increment in low-frequency HRV and DC, respectively. Higher low-frequency HRV and DC, measured during both daytime and nighttime, were associated with decreased hazard of death, but daytime measures showed numerically stronger associations. Results did not substantially vary by zygosity.

CONCLUSIONS: Autonomic inflexibility, and especially vagal withdrawal, are important mechanistic pathways of general mortality risk, independent of familial and genetic factors.

Key Words: autonomic nervous system ■ longitudinal studies ■ death ■ twins

The autonomic nervous system controls basic bodily functions such as heartbeat, digestion, respiration, and blood pressure. Dysregulation of the autonomic system can be a consequence of many diseases or adverse exposures[1–3] and is associated with various pathological conditions, such as high blood pressure, cardiovascular disease (CVD), and death.[4–7] Autonomic function can be measured noninvasively using heart rate variability (HRV), which provides indices of beat-to-beat heart rate fluctuations over time derived from electrocardiographic monitoring data. Reduced HRV is indicative of an imbalance between sympathetic and parasympathetic modulation[8] and has been associated with death primarily in individuals with known CVD.[6,9–11]

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CLINICAL PERSPECTIVE

What Is New?

- In this prospective study of 501 twins, measures of autonomic nervous system dysregulation, including heart rate variability and deceleration capacity (a measure of vagal withdrawal), measured from 24-hour Holter ECGs, were significantly associated with mortality risk.
- The association was independent of familial and genetic factors in addition to traditional cardiovascular risk factors.
- Deceleration capacity exhibited the most robust association with mortality risk when contrasted with conventional metrics of heart rate variability.

What Are the Clinical Implications?

- Ambulatory electrocardiographic metrics of autonomic function, especially deceleration capacity, are important biomarkers of long-term risk of death.
- These measures index the health of a complex cardioneural network that integrates the brain, the autonomic nervous system, and the heart, and whose function is essential for survival.
- The monitoring of autonomic function, and of deceleration capacity in particular, should be evaluated as part of prevention strategies to reduce the risk of adverse health outcomes.

Nonstandard Abbreviations and Acronyms

DC	deceleration capacity
ETS	Emory Twin Study
HRV	heart rate variability
LF	low-frequency
NN	normal-to-normal
VET	Vietnam Era Twin registry

While traditional measures of HRV signal autonomic dysregulation, they do not distinguish between vagal and sympathetic effects.[12] Time domain HRV indices have also been criticized for not being specific to physiological mechanisms.[12] With recent progress in passive sensing technologies, novel indices have been extracted from ECG data. Some of these measures, such as distribution entropy,[13] provide advances in the analysis of complex heart rate fluctuation patterns that may have prognostic significance, but their physiological underpinnings remain unclear.

Deceleration capacity (DC) of heart rate is one such novel metric, but it carries specific physiological significance as a marker of vagal function on the heart, as it indexes the rate of deceleration of the heart rate in short intervals that are consistent with vagal activity.[14,15] This metric, therefore, should allow more direct examination of the relationship of vagal function with health outcomes. Indeed, DC has revealed strong prognostic value in clinical samples with known CVD, with a predictive ability often stronger than conventional HRV metrics.[16–18]

Few studies have evaluated the association of HRV and, specifically, DC, with the risk of death in the general population. Those that have studied community samples have typically examined a limited spectrum of HRV domains.[19,20] In addition, no prior study of death has evaluated daytime and nighttime metrics separately. As autonomic function can be influenced by physical activity and daily stressors during the day, as well as sleep disturbances during the night, examining daytime and nighttime separately may provide incremental predictive information over 24-hour measurements only.

Prior studies suggest that interindividual differences in HRV may be partially explained by genetic factors and familial predisposition, which could also influence the risk of death and cardiac events.[21–23] However, it is unclear to what extent shared genes and familial factors explain the association of HRV with adverse health outcomes. A co-twin control study design provides a natural "counterfactual" design to examine phenotypic associations, as twins are matched for genetic and early familial factors.[24] This design allows the assessment of familial and genetic influences on the associations by comparing the within-pair results with the overall results, and by comparing results in monozygotic and dizygotic twin pairs.

Using a co-twin control design in a sample of male middle-aged veteran twins, we sought to investigate the prospective association of a comprehensive spectrum of HRV frequency domains, as well as of DC, with the risk of death, with all the metrics measured over 24 hours and separating daytime and nighttime HRV. We also evaluated whether familial and genetic factors played a role in the association. We hypothesized that higher values of both daytime and nighttime HRV and DC would be associated with decreased risk of death, and that familial and genetic factors would play a limited role in this association.

METHODS

Data Sharing Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Study Population

The participants in this study were recruited from the VET (Vietnam Era Twin) registry, which is a large national sample of adult male monozygotic and dizygotic twins who served in active duty during the Vietnam war (1964–1975).[25] The present study is based on the 566 twins (283 pairs) who participated in the ETS (Emory Twin Study), a study of the role of biological, psychological, and behavioral risk factors in the development of subclinical CVD.[26,27] ETS included twin pairs who were born between 1946 and 1956, and excluded twin pairs if either member of the twin pair self-reported a history of CVD on the basis of previous survey data obtained by the registry in 1990.[28,29] Twin pairs discordant for depression or posttraumatic stress disorder were oversampled. At the baseline visit, conducted between 2002 and 2010, HRV was measured using 24-hour electrocardiographic monitoring. Of the 566 ETS twins, HRV data were available in 501 twins (225 pairs and 51 singles), which represent the analytical sample for the current study.

At the baseline ETS visit, twin pairs were examined together at Emory University General Clinical Research Center on the same day using identical assessment protocols, which included morning wake-up time, transport, meal timing, and general schedule, to minimize measurement error. All data collection, including ambulatory ECG monitoring, occurred during a 24-hour admission under controlled conditions. Twin brothers maintained an identical schedule; activity was limited to leisurely ambulation within the Emory campus, and all assessment, including the ambulatory ECG monitoring, began and ended at the same time. We obtained twins' comprehensive medical history during a 2-day admission in a clinical research facility and collected anthropometric measurements, behavioral and psychosocial assessments, and autonomic function data using identical protocols. Zygosity was obtained and verified by DNA typing.[30] We obtained written informed consent from all twins, and the Emory University Institutional Review Board approved this research.

Measurements of Autonomic Function

At the baseline ETS visit, each twin wore an ambulatory ECG monitor for 24 hours. We followed previously published procedures to maximize accuracy of recordings and minimize potential confounding.[31] Both twins in the same pair were evaluated at the same time, and their recording times, schedule, and activity level during ECG monitoring were similar. Twins were asked to refrain from smoking and consuming alcohol or coffee during measurements. Each Holter recording was digitally processed and analyzed using methods previously described,[32,33] and was further segmented into daytime (6 AM to 10 PM) and nighttime (10 PM to 6 AM) periods as set by the nursing staff during the overnight stay. The HRV spectrum was computed using an open source, validated toolbox on the edited normal-to-normal (NN) interval time series.[34] A list of NN intervals with annotations denoting normal beats, types of ectopics, and noise was saved and later transferred to an open-source benchmarked toolbox for further processing and analysis with customized software. Before spectral analysis, the NN interval data file was first manually edited to remove nonsinus beats and noise, and any aberrant beats >20% of interval change, nonsinus rhythm, and possible atrial ectopic beats were discarded. We used the Lomb periodogram to interpolate missing beats in the time series, which we found to offer superior performance compared with other interpolation methods.[34] Overall, the proportion of data that were excluded for reasons related to noise and arrhythmia was low (<20% of sample). The heartbeat power spectral analysis was computed over 5-minute sliding windows with 30-second increments over the 24-hour observation period; these measures were then averaged for each patient. If we were unable to evaluate HRV for at least 80% of the day, then we excluded those participants from the analysis. We measured heart rate by calculating the mean of the NN interval time series. We measured the power spectra of the NN time series using standard procedures with the fast Fourier transform. We performed wavelet analysis using the Haar wavelet function to derive the DC from the central part of the phase-rectified signal average. We evaluated 24-hour average, as well as daytime and nighttime average values for 4 discrete frequency bands, including ultra-low-frequency (<0.003 Hz), very-low-frequency (0.0033–0.04 Hz), low-frequency (LF, 0.04–0.15 Hz), and high-frequency (0.15–0.40 Hz).[6,35] The mean DC of heart rate was measured via phase-rectified signal averaging of heart rate decelerations, providing an average speed of heart rate decelerations measured in serial 5-minute windows. The HRV and DC data processing was performed blindly with respect to other twins' characteristics.

Measurements of Mortality Events

Vital status data during follow-up, including dates and causes of deaths, were obtained from the National Death Index database through April 21, 2021. All-cause death was the primary outcome of this study, but we also evaluated CVD-related death in an exploratory analysis, using *International Classification of Diseases, Tenth Revision* (*ICD-10*) codes I21.9, I25, I25.1, I38, I42.9, I50, I60.8, I61.9, and I80.2.

Other Measurements

At the baseline visit, twins received a thorough assessment including medical history and physical

examination by a research nurse or physician assistant. Sociodemographic and anthropometric data, health behaviors, fasting blood glucose, and lipid profile were measured as previously described.[26,28] Habitual physical activity was measured using the Baecke Questionnaire of Habitual Physical Activity, a 16-question instrument assessing physical activity levels at work, during sports and nonsports activities, which are combined in a global physical activity score.[36,37] History of hypertension was defined as systolic blood pressure ≥140 mm Hg or diastolic blood pressure ≥90 mm Hg, or self-reported use of antihypertensive medications, following the Joint National Committee-7 classification, which was the accepted staging of hypertension at the time.[38] History of coronary artery disease that might have occurred from the time of the initial screen in 1990 was also assessed. Diabetes was defined as having a measured fasting glucose of >126 mg/dL or any current treatment with antidiabetic medications. Current use of β blockers, antidepressants, statins, and angiotensin-converting enzyme inhibitors was also recorded. A continuous measure of depressive symptoms was assessed using the Beck Depression Inventory-II score, which includes 21 items each scored from 0 to 3, with a total score ranging from 0 to 63. A clinical diagnosis of major depression and posttraumatic stress disorder (lifetime and current), as well as alcohol abuse disorder, was obtained using the Structured Clinical Interview for *Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition*.

Statistical Analysis

The HRV and DC data were log-transformed owing to nonnormality. We compared the characteristics among twins who died during follow-up with those who survived, using 2 sample *t*-tests for continuous variables and χ^2 tests for categorical variables. The end of follow-up was the last contact date, the date of death, or the last day of available data (April 21, 2021) from the National Death Index, whichever was the latest.

For all analyses, 24-hour average HRV metrics were used as primary exposure of interest, but we also examined daytime and nighttime average HRV metrics separately. To allow comparisons between different autonomic metrics, after log transformation these variables were standardized by computing *z* scores.

Our primary analysis focused on the associations of the within-pair difference in the natural log of HRV and DC as continuous variables with survival time. In a study of twins, within-pair differences intrinsically control for potential confounding by shared genetic and early familial factors. In our study, within-pair comparisons also controlled for environmental factors during ambulatory monitoring, because twins were examined together and followed a similar schedule on the testing day. The within-pair effects were calculated as the departure of each twin from the pair average for each metric as a continuous variable.[39]

To allow an evaluation of potential familial influence on the associations of interest, within-pair analyses were preceded by analyses that treated twins as individuals. If the within-pair estimates are attenuated compared with the overall results, then familial factors play a role. First, Kaplan–Meier curves for all-cause death were computed in the overall sample of 501 twins, with the natural log of each autonomic metric dichotomized at the median value. Next, Kaplan–Meier curves were recalculated among 225 complete pairs (n=450), to examine within-pair differences in log-transformed metrics. In the within-pair analyses, twins with a higher 24-hour log-HRV or DC (a within-pair difference>0) were compared with their brothers with a lower value (a within-pair difference <0). Log-rank tests were used to compare survival curves.

For multivariable analyses, we fitted frailty models that accounted for twin pair as random effect. The frailty model is an extension of the Cox proportional hazard model, with random effects to account for heterogeneity in clustered data (such as twin pairs).[40] As above, initially we conducted analyses in twins as individuals, and then examined within-pair differences among complete pairs. In within-pair analysis, the β coefficients represent the hazard ratios (HRs) for all-cause death per 1-SD increment in within-pair difference in log-HRV and log-DC metrics, comparing twins with higher values to their brothers with lower values.

To avoid model overfitting, we constructed a series of models to examine the impact of sets of a priori selected variables on the association of interest. The base model, or model 1, was unadjusted. We then progressively adjusted for sociodemographic and behavioral variables (education, employment status, ever-smoking status, history of alcohol abuse, and physical activity) in model 2, and further adjusted for risk factors that are likely related to both HRV and mortality outcomes (body mass index, history of hypertension, history of coronary artery disease, and diabetes) in model 3.[41] In model 4, we additionally adjusted for medication use, including medications most likely to affect autonomic function (β blockers and antidepressants).

To assess potential shared genetic influence on autonomic metrics and adverse health outcome, we evaluated effect modification by zygosity. Because monozygotic twin pairs share 100% of their genes while dizygotic twin pairs share only 50% on average, if a larger effect of HRV on adverse health outcomes is observed within dizygotic pairs than in monozygotic pairs, then it may suggest that genetic factors play a role in this association.

We conducted additional analyses as an expansion of our primary analysis. First, we examined whether the results remained robust after additionally adjusting for lifetime history of depression and posttraumatic stress disorder, as well as adjusting for 24-hour average heart rate, as prior research pointed out that the relationship between HRV and death could be partially attributable to concurrent changes in heart rate.[42] Second, we evaluated the association between baseline HRV and time to CVD-specific death during follow-up.

Missing data were rare (<5%) for all variables; thus, we used all available data without imputation. We checked linearity assumptions of all continuous variables, as well as potential multicollinearity by variance inflation factors. A 2-sided *P* value of <0.05 was used to indicate statistical significance, and HRs and associated 95% CIs were calculated for model parameters. All statistical analyses were performed using SAS version 9.4 (SAS Institute, Cary, NC) and Stata 14.0 (StataCorp, College Station, TX).

RESULTS

Participants' Characteristics

Of the 501 twins in the total sample, 483 (96%) were White participants, with a mean age (SD) of 56 (3) years (Table 1). During a mean (SD) follow-up of 12 (3) years, a total of 77 twins died (15%). There were 55 discordant pairs for death; the remaining 18 deaths occurred in the same 9 twin pairs. Comparing deceased twins with those who survived, twins who died were more likely to be smokers, were less physically active, and had lower HRV in the very-low-frequency and LF domains, and lower DC (Table 1).

HRV and Death

The mean (SD) time to death of the 77 twins who died was 8.9 (SD, 5.1) years. Of these deaths, 22 (29%) were due to cancer and 14 (18%) to CVD causes. Other causes of death included motor vehicle accident; suicide; and endocrine, lung, or gastrointestinal disorders.

Kaplan–Meier survival curves in the overall population showed that twins who had a higher (median or above) 24-hour average LF HRV and DC at baseline had significantly better survival than those with lower values (below the median) (Figure 1). When compared with their brothers in within-pair analysis, these results remained consistent (Figure 2). In contrast, twins with higher ultra-low-frequency, very-low-frequency, and high-frequency 24-hour HRV exhibited smaller and nonsignificant differences in survival, both in the overall sample and when compared with their brothers with lower corresponding metrics.

In multivariable analysis, higher values for 24-hour HRV showed HRs for death that were less than 1 across almost all models, indicating reduced risk for all-cause mortality. However, the association was consistently significant only for LF HRV and DC, both in the overall sample (Table 2) and when comparing brothers within pairs (Table 3). In unadjusted within-pair analysis, there was a 21% and 27% decreased risk per 1-SD increment in log-LF HRV and log-DC, respectively. This association was not meaningfully altered after adjusting for sociodemographic and behavioral factors, CVD risk factors, and medication use. In the full model, there was a 22% and 27% lower risk of death per 1-SD increment in log-LF HRV and log-DC, respectively. Daytime HRV showed slightly stronger associations for LF and DC compared with nighttime values (Tables 2 and 3). Specifically, comparing twins within pairs, a 1-SD increment in daytime log-LF HRV and log-DC showed significantly decreased risk for all-cause death, with fully adjusted HRs of 0.77 (95% CI, 0.61–0.98) and 0.71 (95% CI, 0.55–0.92) for LF HRV and DC, respectively. The corresponding estimates for nighttime LF HRV and DC were somewhat weaker and not consistently significant.

Overall, the associations of HRV and DC metrics with death were similar in dizygotic and monozygotic twins across virtually all models, and none of the interaction terms with zygosity were significant (Table S1).

Additional Analyses

Adjustment for lifetime history of depression and posttraumatic stress disorder, and for 24-hour average heart rate, did not materially change the associations in any of the models (Table S2). Additional adjustment for DC in the LF models, or adjustment for LF in the DC models, did not substantially change the results, demonstrating that LF and DC were largely independent of each other in their associations with all-cause death (Table S3).

There were only 14 CVD-specific deaths. None of the baseline autonomic metrics were significantly associated with the risk of CVD-specific death in any of the models, although effect sizes, especially for DC, were similar to those of all-cause death (Table S4). Stratified analysis by zygosity was not completed for CVD death, as the number of events was too small to yield reliable estimates.

DISCUSSION

In this co-twin control study, a 1-SD increment in LF HRV and in DC, denoting better autonomic flexibility, was associated with 21% and 27% decreased hazard for all-cause death during an average of 12 years' follow-up. The associations remained robust after adjusting for sociodemographic, behavioral, and health-related factors, and medication use. Among

Table 1. Characteristics of 501 Twins With Available HRV Data in ETS

Characteristics, mean (SD)	Total (N=501)	Died (n=77)	Survived (n=424)	*P* value
Sociodemographic factors				
Age, y	56 (3)	55 (3)	56 (3)	0.428
White race, n (%)	483 (96)	71 (92)	412 (97)	0.031
Years of education	15 (2)	15 (2)	15 (2)	0.787
Employed, n (%)	404 (81)	50 (65)	354 (83)	<0.001
Health factors				
BMI	30 (5)	29 (6)	30 (5)	0.332
Ever smokers, n (%)	330 (66)	59 (77)	271 (64)	0.030
Alcohol abuse, n (%)	234 (47)	40 (52)	194 (46)	0.316
Baecke score for physical activity	7.3 (1.8)	6.5 (2.1)	7.4 (1.7)	<0.001
Systolic blood pressure, mm Hg	124 (10)	121 (10)	124 (10)	0.022
Diastolic blood pressure, mm Hg	74 (9)	73 (9)	74 (8)	0.376
History of hypertension, n (%)	187 (37)	32 (42)	155 (37)	0.412
History of diabetes, n (%)	62 (12)	15 (19)	47 (11)	0.040
Prior history of CAD, n (%)	55 (11)	14 (18)	41 (10)	0.028
Lifetime history of PTSD, n (%)	74 (15)	14 (18)	60 (14)	0.359
Lifetime history of depression, n (%)	137 (27)	27 (35)	110 (26)	0.099
Current PTSD, n (%)	33 (7)	9 (12)	24 (6)	0.050
Current depression, n (%)	16 (3)	5 (6)	11 (3)	0.073
BDI-II score	6.0 (7.7)	8.5 (9.5)	5.6 (7.3)	0.003
Medication use				
β Blockers, n (%)	41 (8)	10 (13)	31 (7)	0.095
Antidepressants, n (%)	79 (16)	23 (30)	56 (13)	<0.001
Statin, n (%)	130 (26)	20 (26)	110 (26)	0.995
ACE inhibitor, n (%)	79 (16)	13 (17)	66 (16)	0.770
Zygosity				0.392
Monozygotic, n (%)	310 (62)	51 (66)	259 (61)	
Dizygotic, n (%)	191 (38)	26 (34)	165 (39)	
Heart rate variability				
24-h average				
ln ULF	6.6 (1.0)	6.5 (0.8)	6.6 (1.0)	0.254
ln VLF	7.6 (0.9)	7.5 (0.8)	7.7 (0.9)	0.051
ln LF	6.6 (0.9)	6.4 (0.9)	6.7 (0.8)	0.001
ln HF	5.5 (0.9)	5.4 (0.8)	5.5 (0.9)	0.601
ln DC	2.3 (0.4)	2.2 (0.4)	2.4 (0.3)	0.002
Daytime average				
ln ULF	6.7 (1.0)	6.6 (0.9)	6.8 (1.0)	0.150
ln VLF	7.6 (0.9)	7.4 (0.9)	7.7 (0.9)	0.023
ln LF	6.6 (0.9)	6.3 (0.9)	6.7 (0.9)	<0.001
ln HF	5.4 (0.9)	5.3 (0.9)	5.4 (0.9)	0.249
ln DC	2.3 (0.4)	2.2 (0.4)	2.3 (0.4)	<0.001
Nighttime average				
ln ULF	6.3 (1.3)	6.1 (1.3)	6.3 (1.2)	0.161
ln VLF	7.7 (1.0)	7.4 (1.2)	7.7 (1.1)	0.048
ln LF	6.6 (1.0)	6.4 (1.1)	6.7 (1.0)	0.010
ln HF	5.6 (1.0)	5.6 (1.0)	5.6 (1.0)	0.924
ln DC	2.4 (0.4)	2.3 (0.5)	2.5 (0.4)	0.017

ACE indicates angiotensin-converting enzyme; BDI, Beck Depression Inventory; BMI, body mass index; CAD, coronary artery disease; DC, deceleration capacity; ETS, Emory Twin Study; HF, high frequency; HRV, heart rate variability; LF, low frequency; PTSD, posttraumatic stress disorder; SD, standard deviation; ULF, ultra-low frequency; and VLF, very low frequency.

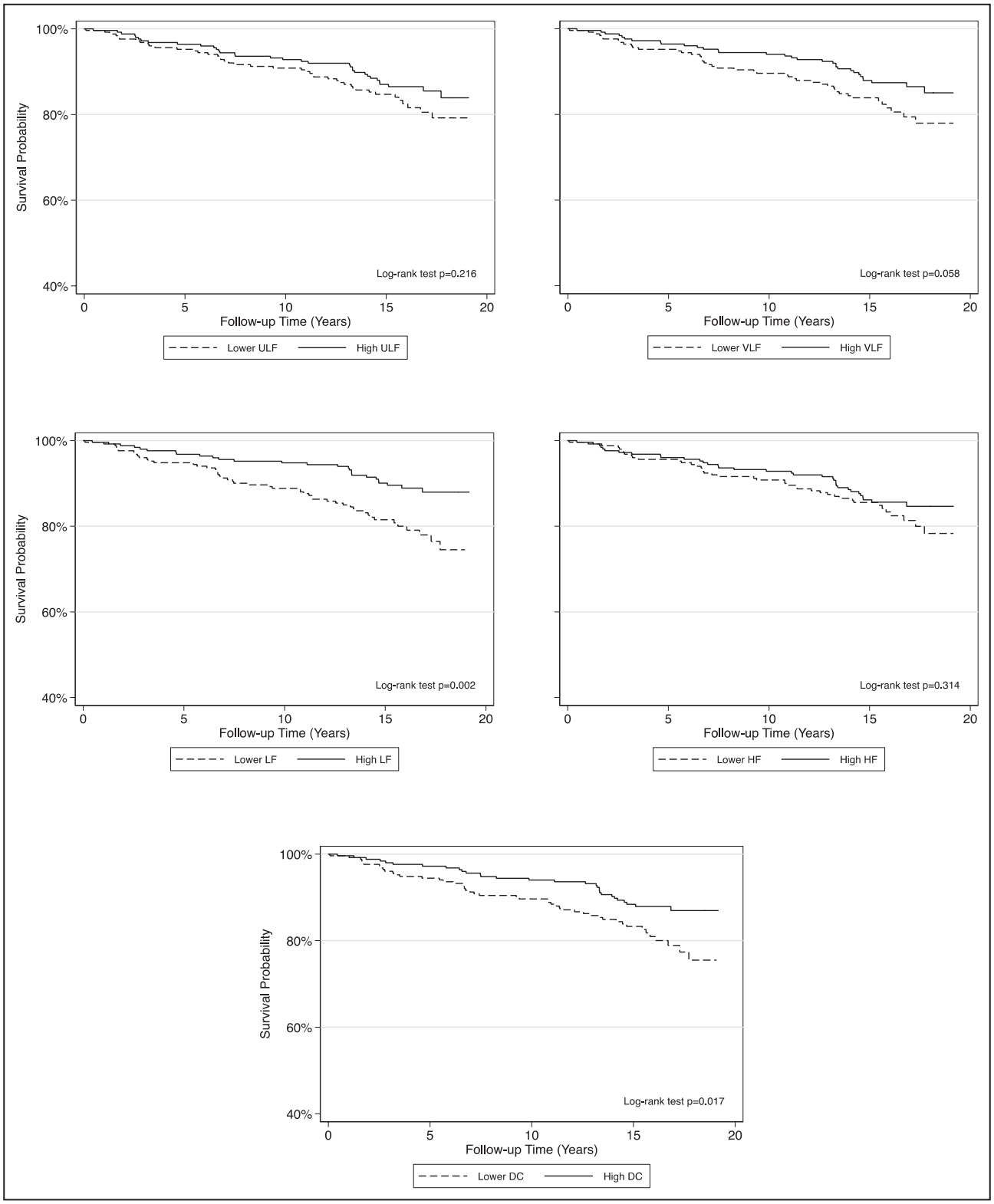

Figure 1. Kaplan–Meier survival probabilities for all-cause death by levels of 24-hour average HRV domains in the overall sample (n=501).

HRV metrics were dichotomized at the median value. Curves compare twins with lower HRV (below the median) with twins with high HRV (median or above). DC indicates deceleration capacity; HF, high frequency; HRV, heart rate variability; ULF, ultra-low frequency; and VLF, very low frequency.

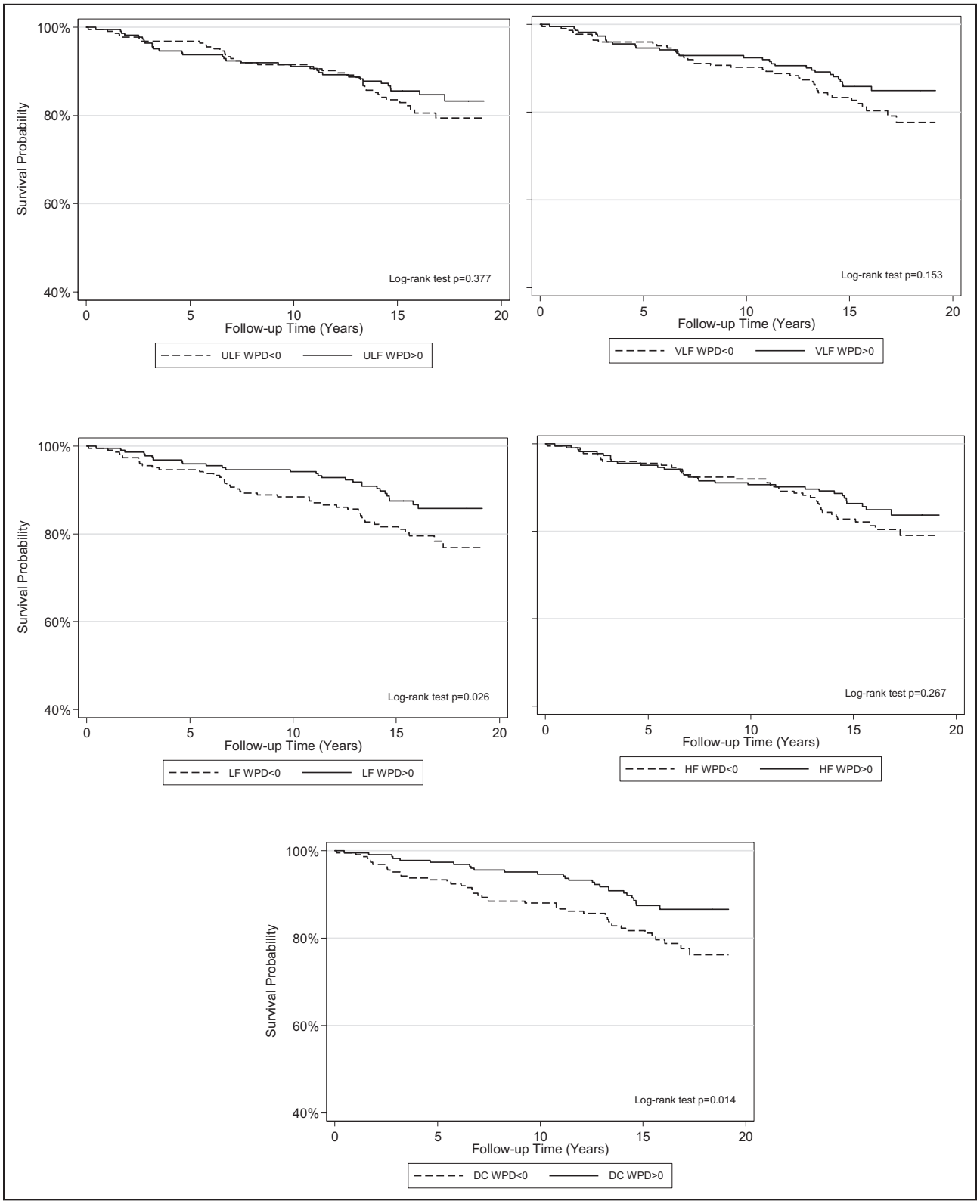

Figure 2. Kaplan–Meier survival probabilities for all-cause death by within-pair difference of 24-hour average HRV domains (n=450).

Twins with a higher 24-hour average HRV value than their brothers (within-pair difference>0) were compared with their co-twins (within-pair difference<0). DC indicates deceleration capacity; HF, high frequency; HRV, heart rate variability; LF, low frequency; ULF, ultra-low frequency; VLF, very low frequency; and WPD, within-pair difference.

Table 2. Individual Twin Analysis of the Association Between Baseline HRV and Time to All-Cause Death During Follow-Up

	Model 1*, HR (95% CI)	Model 2†, HR (95% CI)	Model 3‡, HR (95% CI)	Model 4§, HR (95% CI)
24-h average HRV (n=501)				
ln ULF	0.89 (0.75–1.05)	0.92 (0.76–1.12)	0.94 (0.77–1.14)	0.94 (0.77–1.15)
ln VLF	0.83 (0.71–0.98)‖	0.87 (0.73–1.05)	0.89 (0.74–1.07)	0.89 (0.73–1.08)
ln LF	0.75 (0.63–0.88)‖	0.79 (0.66–0.95)‖	0.79 (0.66–0.96)‖	0.80 (0.66–0.96)‖
ln HF	0.92 (0.74–1.15)	1.00 (0.78–1.28)	1.00 (0.78–1.29)	1.01 (0.79–1.30)
ln DC	0.67 (0.54–0.84)‖	0.77 (0.61–0.97)‖	0.75 (0.59–0.96)‖	0.76 (0.59–0.97)‖
Daytime average HRV (n=494)				
ln ULF	0.87 (0.74–1.02)	0.89 (0.75–1.06)	0.91 (0.76–1.08)	0.91 (0.76–1.09)
ln VLF	0.82 (0.71–0.96)‖	0.85 (0.72–1.01)	0.87 (0.73–1.03)	0.87 (0.73–1.04)
ln LF	0.74 (0.63–0.86)‖	0.78 (0.66–0.92)‖	0.78 (0.66–0.93)‖	0.78 (0.65–0.93)‖
ln HF	0.86 (0.69–1.06)	0.91 (0.73–1.15)	0.92 (0.73–1.15)	0.92 (0.73–1.16)
ln DC	0.64 (0.52–0.80)‖	0.72 (0.57–0.91)‖	0.69 (0.54–0.88)‖	0.69 (0.54–0.89)‖
Nighttime average HRV (n=466)				
ln ULF	0.87 (0.73–1.05)	0.90 (0.74–1.09)	0.91 (0.75–1.11)	0.92 (0.75–1.12)
ln VLF	0.83 (0.70–0.98)‖	0.87 (0.72–1.04)	0.88 (0.73–1.07)	0.89 (0.73–1.08)
ln LF	0.78 (0.65–0.93)‖	0.83 (0.68–1.01)	0.84 (0.69–1.02)	0.84 (0.69–1.03)
ln HF	0.98 (0.78–1.24)	1.08 (0.83–1.39)	1.10 (0.84–1.43)	1.11 (0.85–1.44)
ln DC	0.72 (0.57–0.91)‖	0.85 (0.66–1.09)	0.86 (0.67–1.10)	0.86 (0.67–1.11)

There were 77 total deaths. Results are shown as standardized HRs in the multivariable Cox frailty models per 1-SD difference in log-HRV. BMI indicates body mass index; DC, deceleration capacity; HF, high frequency; HR, hazard ratio; HRV, heart rate variability; LF, low frequency; ULF, ultra-low frequency; and VLF, very low frequency.

*Base model was unadjusted.

†Model 2=model 1+sociodemographic and behavioral factors, including education, employment status, ever smoking status, alcohol abuse, and physical activity.

‡Model 3=model 2+BMI, history of hypertension, history of coronary artery disease, and diabetes.

§Model 4=model 3+β blockers and antidepressants.

‖Indicates significant association at $P<0.05$.

HRV parameters, LF HRV and DC showed the strongest and most consistent associations with all-cause death. Overall, daytime HRV metrics showed slightly stronger associations than nighttime HRV. Because the association was found within twin pairs, and results did not substantially vary when analyzing twins as individuals and within pairs, familial factors may not play a confounding role in these associations. Genetic background also did not play a role since results were similar by zygosity.

Our findings are consistent with a meta-analysis of 28 cohort studies in patients with known CVD, showing that individuals with a lower HRV had 112% higher risk of all-cause death.[9] Our results also agree with community-based research in middle-aged or elderly participants from decades ago,[5,19,43–45] suggesting that reduced HRV is associated with an elevated risk of death, which could not be attributable to a specific cause. Compared with these prior studies, our investigation was able to control for familiar factors by comparing HRV-discordant twins within pairs. Furthermore, in contrast with most previous community reports, our study examined a comprehensive spectrum of autonomic measures that included the novel DC metric in addition to several HRV domains, all assessed over 24 hours as well as separately in daytime and nighttime segments.

Our investigation is one of the first to examine DC as a predictor of death in a community cohort. In our study, DC exhibited the most robust association with mortality risk when contrasted with conventional HRV metrics. DC is a measure of cardiac vagal modulation, indexing flexibility of heart rate in response to vagal influences.[16] Prior experimental and clinical studies have demonstrated a cardioprotective role of vagal activity.[46–48] Consistent with these experimental data, studies of patients with CVD have linked higher values of DC to lower all-cause death, with a protective effect that was of a larger magnitude than conventional HRV metrics.[16–18] Our findings agree with and expand these previous data from selected samples of patients with CVD and support DC as a promising biomarker of healthy autonomic function.

In line with prior investigations,[19,49] we found that LF power displayed robust associations with death. The power in the LF HRV domain is in part modulated through the sympathetic nervous system as a response to oscillations in blood pressure,[50] and reduced LF power may indicate severe autonomic dysregulation.[49,51] Contrary to previous studies, however, we did

Table 3. Within-Pair Analysis of the Association Between Baseline HRV and Time to All-Cause Death During Follow-Up

	Model 1*, HR (95% CI)	Model 2†, HR (95% CI)	Model 3‡, HR (95% CI)	Model 4§, HR (95% CI)
24-h average HRV (n=450)				
ln ULF	0.90 (0.72–1.11)	0.89 (0.70–1.14)	0.90 (0.71–1.14)	0.90 (0.71–1.14)
ln VLF	0.85 (0.68–1.05)	0.84 (0.66–1.06)	0.85 (0.68–1.07)	0.85 (0.67–1.07)
ln LF	0.79 (0.63–0.98)‖	0.79 (0.62–0.99)‖	0.79 (0.63–0.98)‖	0.78 (0.62–0.98)‖
ln HF	0.95 (0.72–1.26)	0.96 (0.72–1.28)	0.96 (0.72–1.28)	0.96 (0.72–1.28)
ln DC	0.73 (0.56–0.94)‖	0.75 (0.58–0.97)‖	0.74 (0.57–0.96)‖	0.73 (0.56–0.95)‖
Daytime average HRV (n=444)				
ln ULF	0.90 (0.73–1.10)	0.89 (0.71–1.13)	0.90 (0.72–1.13)	0.90 (0.71–1.13)
ln VLF	0.86 (0.70–1.06)	0.85 (0.68–1.06)	0.86 (0.68–1.07)	0.85 (0.68–1.07)
ln LF	0.79 (0.63–0.98)‖	0.78 (0.62–0.99)‖	0.78 (0.62–0.98)‖	0.77 (0.61–0.98)‖
ln HF	0.89 (0.67–1.17)	0.89 (0.67–1.18)	0.89 (0.67–1.18)	0.88 (0.66–1.17)
ln DC	0.72 (0.56–0.94)‖	0.74 (0.57–0.96)‖	0.73 (0.57–0.94)‖	0.71 (0.55–0.92)‖
Nighttime average HRV (n=418)				
ln ULF	0.95 (0.77–1.18)	0.94 (0.74–1.20)	0.93 (0.73–1.17)	0.91 (0.72–1.16)
ln VLF	0.89 (0.71–1.10)	0.87 (0.68–1.11)	0.85 (0.67–1.09)	0.84 (0.65–1.08)
ln LF	0.82 (0.64–1.04)	0.81 (0.63–1.04)	0.79 (0.61–1.02)	0.77 (0.59–1.00)
ln HF	1.05 (0.79–1.40)	1.09 (0.81–1.47)	1.08 (0.80–1.46)	1.06 (0.78–1.44)
ln DC	0.75 (0.57–0.98)‖	0.79 (0.61–1.04)	0.79 (0.61–1.02)	0.76 (0.58–0.99)‖

There were 55 discordant pairs for mortality in the within-pair analysis. Results are shown as standardized HRs in the multivariable Cox frailty models, per 1-SD within-pair difference in log-HRV, comparing twins with higher HRV to their brothers with lower HRV. BMI indicates body mass index; DC, deceleration capacity; HF, high frequency; HR, hazard ratio; HRV, heart rate variability; LF, low frequency; ULF, ultra-low frequency; and VLF, very low frequency.

*Base model was unadjusted.

†Model 2=model 1+sociodemographic and behavioral factors, including education, employment status, ever-smoking status, alcohol abuse, and physical activity.

‡Model 3=model 2+BMI, history of hypertension, history of coronary artery disease, and diabetes.

§Model 4=model 3+β blockers and antidepressants.

‖Indicates significant association at $P<0.05$.

not find significant associations of very-low-frequency and high-frequency HRV with all-cause death.[19] This discrepancy of results may stem from differences in study design, participant characteristics, or length of follow-up. Moreover, in contrast with other studies,[10,52] we did not find a significant association between HRV and CVD death. However, the number of CVD deaths was small in our study, and the cause-specific death information was based on death certificate codes only.

Because variations in exposures and activity can influence the measurement of HRV during the day, nighttime HRV assessed during sleep could be a more precise indicator of autonomic regulation. During sleep, gross physical activity and postural effects are minimized, as are other potential influences such as meals, fluid intake, and psychosocial exposures.[53] On the other hand, nighttime HRV could be confounded by sleep quality or sleep disorders, which may have an impact on the magnitude of the association between HRV and death. Furthermore, the daytime may have more stressful exposures that can uncover underlying vulnerabilities through HRV, while the nighttime is a more controlled period from an autonomic system standpoint, without as much autonomic stimulation except for sleep disturbance. To date, no prior study has evaluated daytime and nighttime HRV for their association with mortality risk. We found that both daytime and nighttime HRV are associated with risk for all-cause death, but daytime HRV tended to show stronger associations than nighttime HRV. This was especially noted for DC, which is predominantly modulated by the parasympathetic nervous system. These data support the notion that alterations in daytime parasympathetic function carry important information for mortality risk.

The pathophysiological mechanisms linking HRV and DC with risk of death remain unclear. Autonomic function indexed by HRV reflects overall sympathovagal balance and autonomic flexibility,[54] while DC is a more specific metric of vagal modulation.[16] These measures signal the health of a complex cardioneural network that integrates the brain, the autonomic nervous system, and the heart, and whose function is essential for survival.[55] Furthermore, both the sympathetic and the parasympathetic nervous systems participate in the adaptive regulation of the inflammatory response, which is implicated in many disease processes. The vagus, in particular, is involved in the regulation of the inflammatory reflex through the cholinergic anti-inflammatory pathway,[56] and HRV metrics indexing vagal function have been inversely associated

with inflammatory biomarkers.[57] Thus, autonomic dysregulation leading to inflammation may affect the function of virtually all organ systems and therefore overall survival. It is also possible that the indices of autonomic dysregulation in our study are markers of unfavorable general health, in part through subclinical disease. Irrespective of the underlying mechanisms, our results support the concept that ambulatory ECG metrics of autonomic function, especially DC and LF HRV, are important biomarkers of healthy autonomic function and long-term all-cause mortality risk.

A limitation of our study is the small number of CVD deaths, with resulting limited power to detect statistically significant associations in the CVD mortality analysis. This may explain why the effect sizes, especially for DC, were large, but the associations were not statistically significant. Second, the inclusion of only male and mostly White middle-aged adults and their unique military training experience and physical health status may be different from the general population, which may limit generalizability. However, our co-twin control study design should maximize internal validity and precision by intrinsically adjusting for unknown or unmeasured familial and environmental confounders. This is also the first investigation to examine the association of DC, along with a full spectrum of HRV frequency domains, over 24 hours as well as during daytime and nighttime, with mortality outcomes.

CONCLUSIONS

In the context of a twin design, the present study provides evidence for autonomic regulation as an important factor in mortality risk. Our data underscore DC, a robust parasympathetic metric, as an important biomarker for risk of death. Shared familial factors do not appear to play a major role in these associations. Our study also suggests that 24-hour or nighttime measures of HRV do not provide an advantage to daytime assessments only. There is a critical need to evaluate whether the monitoring of autonomic function, and of DC in particular, can be useful as part of prevention strategies to reduce the risk of adverse health outcomes. Interventions to ameliorate autonomic dysfunction should also be tested in the future.

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Disclosures

None.

Supplemental Material

Tables S1–S4

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