Chronic kidney disease

Thio, C. H. L.

DOI:
10.33612/diss.133648108

Heart rate variability and its relation to chronic kidney disease: Results from the PREVEND Study

Christian HL Thio, Arie M van Roon, Joop D Lefrandt, Ron T Gansevoort, and Harold Snieder

Psychosom Med. 2018;80(3):307–16.

CHAPTER

3

ABSTRACT

Objective. In the general population, reduced heart rate variability (HRV) has been associated with cardiovascular disease. However, its relation to chronic kidney disease (CKD) is debated. We therefore investigated the relation between low HRV and renal outcomes.

Methods. In the population-based PREVEND Study, renal outcomes (CKD, eGFR, urinary albumin) were measured at baseline and three consecutive examinations. HRV measures (among which SDNN, standard deviation of normal-to-normal RR-intervals) were calculated from time-series of beat-to-beat pulse-wave recordings at baseline. The lowest (risk) quartile was compared to the upper three quartiles combined, in multivariable survival and linear mixed-effects analyses.

Results. In 4605 participants (49% males, age range 33-80, 0.6% blacks), we observed 341 new cases of CKD during a median follow-up duration of 7.4 years. Low SDNN was associated with higher incidence of CKD (crude HR: 1.66, 95%CI [1.30;2.12], p<0.001), but this association was no longer significant after adjustment for age, sex, and cardiovascular risk factors (adjusted HR: 1.13, 95%CI [0.86;1.48], p=0.40, similar for other HRV measures). No associations between SDNN and eGFR trajectories were found in the total sample. However, in a subgroup of participants with baseline CKD (N=939), we found a significant association of low SDNN (but not other HRV measures) with lower baseline eGFR, even after multivariable adjustment (adjusted $\beta_{\text{level difference}}$=-3.73 ml/min/1.73m^2, 95%CI [-6.70;-0.75], p=0.014), but not with steeper eGFR decline.

Conclusions. These results suggest that reduced HRV may be a complication of CKD rather than a causal factor.

LIST OF ABBREVIATIONS

CKD = chronic kidney disease
eGFR = estimated glomerular filtration rate
HF = high frequency power
HRV = heart rate variability
LF = low frequency power
PREVEND Study= Prevention of REnal and Vascular ENdstage Disease Study
rMSSD = root mean square of successive differences
SDNN = standard deviation of normal-to-normal RR-intervals
UAE = urinary albumin excretion

BACKGROUND

Chronic kidney disease (CKD) is a group of heterogeneous disorders characterized by kidney damage and impaired renal function, and is defined by an elevated urinary albumin excretion (UAE), a decreased glomerular filtration rate (GFR), or a combination of both.[1-3] The most important risk factors for CKD are diabetes and hypertension. However, it has been observed that CKD can also occur in the absence of these risk factors.[4,5] This suggests that other mechanisms may be involved in the development of CKD.

A potential causal mechanism involves imbalance of the autonomic nervous system, in which parasympathetic function is decreased relative to sympathetic function. Hypothetically, autonomic imbalance causes renal damage through changes in renal hemodynamics. In animal studies, stimulation of renal sympathetic afferents affected renal hemodynamics, while renal (sympathetic) denervation in these animals attenuated progression of kidney failure.[6-8] In humans, a non-invasive way of assessing autonomic function is by calculating heart rate variability (HRV), a measure of autonomic control over heart rate. It is the variation in duration between normal-to-normal (NN) RR-intervals.[9-12] Moderate-to-high HRV indicates healthy autonomic function, while low HRV reflects poor autonomic function, and has been associated with cardiovascular risk factors and adverse cardiovascular outcomes.[10,11,13-16] The relation between HRV and CKD has been explored in several small-scale studies. Participants with CKD were found to have lower HRV compared to those without CKD. Also, low HRV was associated with adverse outcomes during follow-up (i.e. progression to end-stage renal disease and mortality) in CKD patients, although results are inconsistent between studies.[17-23] The mechanisms underlying this association are still under investigation, but it is commonly believed that autonomic imbalance is a complication of renal damage[24].

However, in the Atherosclerosis Risk in Communities (ARIC)-cohort, a 20-108% higher incidence of CKD-related hospitalization and/or end-stage renal disease (ESRD) was observed in those with low HRV (first quartile) compared to those with normal-to-high HRV (upper three quartiles combined), even in participants with normal kidney function at baseline.[25] This suggests that autonomic imbalance may also play a role in the pathophysiology of CKD. To our knowledge, this finding has not yet been verified in other population-based longitudinal studies. If

autonomic imbalance is identified as a mechanism of renal damage, this may lead to improved risk prediction and novel therapeutic options.

In this study, our primary aim was therefore to investigate the association between HRV and new-onset CKD in a sample of the general population. Furthermore, we assessed whether low HRV was associated with baseline levels of eGFR and UAE and change in these parameters during follow-up.

METHODS

Study sample and design

We used data from the Prevention of REnal and Vascular ENdstage Disease (PREVEND) cohort study. Details of this study have been described elsewhere.26 In brief, 8592 individuals, sampled from the general population of Groningen, the Netherlands, completed an extensive examination between 1997- 1998. The second, third, fourth and fifth examination were completed in 2003, 2006, 2008 and 2012, respectively. For the present study, we refer to the second examination as 'baseline', as this was the first examination that included additional beat-to-beat blood pressure recordings that were used for calculation of HRV parameters. This examination was attended by 6894 participants, of which 2289 had missing HRV measures (due to either technical failure (N=397) or due to poor quality signal or excessive amount of artifacts in the recording (N=1892)), leaving 4605 participants for the present analyses. All participants gave written informed consent. The PREVEND Study was approved by the medical ethics committee of the University Medical Center Groningen, and conducted in accordance with the Helsinki Declaration guidelines.

Measurement

HRV measures

Details of the HRV measurement procedure in the PREVEND study have been described previously [27]. In brief, participants were measured in a supine position, in a quiet room kept at a constant temperature of 22°C. Participants were not allowed to talk or move during the procedure. Beat-to-beat heart rate was assessed by non-invasive 15-min pulse wave measurement using a Portapres® device (FMS Finapres Medical systems BV, Amsterdam, The Netherlands)[28] at baseline. From these 15-min measurements, we selected the last 4 to 5 minutes of stationary time-series of pulse wave data. Using CARSPAN v2.0 software [29], these time-

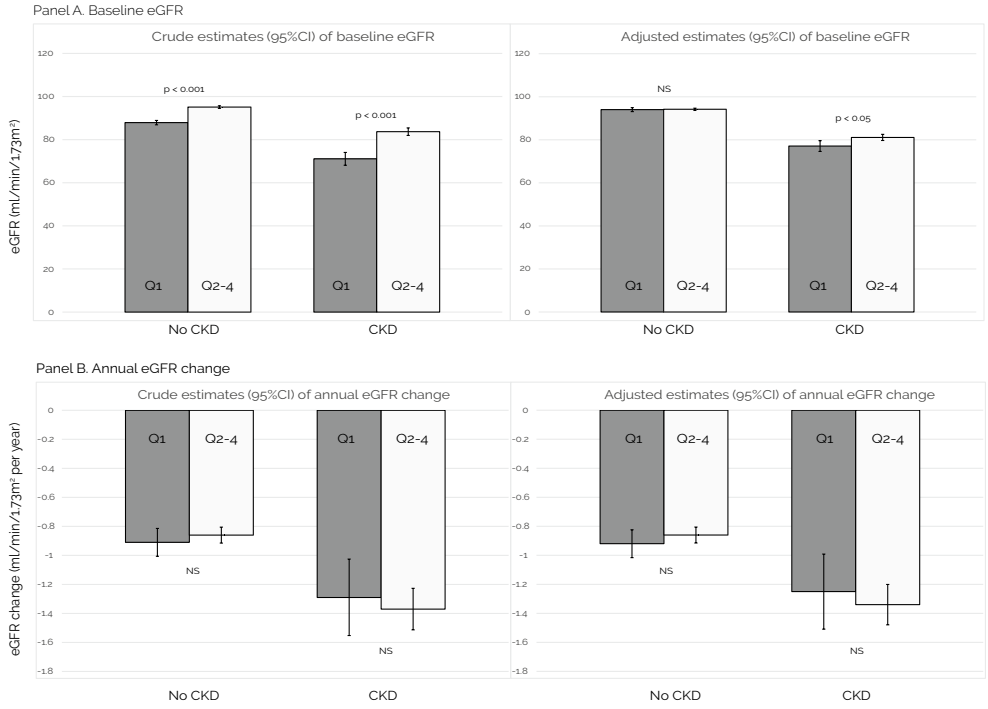
series were visually pre-processed to exclude cardiac arrhythmias, artefacts, electrical 'noise', or aberrant beats. Normal-to-normal RR-intervals from the beat-to-beat pulse wave signals were detected with an accuracy of 5ms (sampling frequency of 200 Hz). Artifacts were removed and the resulting gaps interpolated as described previously[30]. After pre-processing, HRV measures were calculated using the same CARSPAN software. HRV measures included standard deviation of normal-to-normal RR-intervals (SDNN) and root mean square of successive differences between normal-to-normal RR-intervals (rMSSD). To quantify cyclic changes in heart rate, we calculated high frequency (HF) and low frequency (LF) power (area under the power spectral density curve) by Fourier spectral analysis, and the ratio between LF/HF. LF power was defined as the total area between 0.04 and 0.15Hz, and HF power was defined as the total area between 0.15 and 0.40Hz[9-12]. HRV was categorized into low (lowest quartile, Q1) and moderate-to-high (upper three quartiles combined, Q2-4) to allow direct comparison to the work of Brotman et al[25].

Renal outcomes

Details of the assessment of eGFR and UAE have been described elsewhere[31]. In brief, participants collected two consecutive 24h-urine specimens at each screening round. The collected urine was stored cold (4°C) for a maximum of four days before handing it in. After this, urine specimens were stored at -20°C. Furthermore, fasting blood samples were obtained and stored at -80°C.

Measurement of serum creatinine was performed by an enzymatic method on a Roche Modular analyzer using reagents and calibrators from Roche (Roche Diagnostics, Mannheim, Germany), with intra- and interassay coefficients of variation of 0.9% and 2.9%, respectively. Serum cystatin C concentration was measured by a Gentian cystatin C Immunoassay (Gentian AS Moss, Norway) on a Modular analyzer (Roche Diagnostics). Cystatin C was calibrated directly using the standard supplied by the manufacturer (traceable to the International Federation of Clinical Chemistry Working Group for Standardization of Serum Cystatin C)[32]. The intra- and interassay coefficients of variation were <4.1% and <3.3%, respectively. Urinary albumin concentration (UAC) was measured by nephelometry with a lower threshold of detection of 2.3mg/L, and intra- and interassay coefficient of variation of 2.2% and 2.6%, respectively (Dade Behring Diagnostic, Marburg, Germany). UAC was multiplied by urine volume to obtain a value of UAE in mg/24h. The two 24h

Figure 1. Associations of SDNN (Q1 vs Q2-4) with baseline eGFR level (panel A) and annual change in eGFR (panel B).



SDNN: standard deviation of normal-to-normal RR intervals; eGFR: estimated glomerular filtration rate; CKD: chronic kidney disease; NS: non-significant (p > 0.05). Covariates were centered to obtain adjusted estimates. Due to centering, estimates may differ slightly from Table 4.

UAE values of each subject per examination were averaged. eGFR was calculated according to the 2012 CKD-EPI creatinine-cystatin C equation[33]. CKD was defined as an eGFR<60ml/min/1.73m², a UAE≥30mg/24h, or both, according to the 2011 revised KDIGO guidelines[2].

Covariates

Known cardiovascular risk factors were included as covariates and assessed at baseline. Body-mass index (BMI: weight/height²) and waist-hip circumference ratio (WHR) were calculated from anthropometrics. Mean inter-beat interval (IBI) was calculated from time-series of beat-to-beat heart rate data. Smoking status was defined as self-reported never, former, or current smoker (subdivided in <6 cigarettes, 6-20 cigarettes, and >20 cigarettes daily). History of cardiovascular disease (CVD) was assessed using questionnaires, and was defined as a history of any cardio- or cerebro-vascular events. Hypertension was defined as SBP≥140mmHg, DBP≥90mmHg, or self-reported or pharmacy-reported prescribed use of blood pressure-lowering drugs, including ACE-inhibitors,

angiotensin-II receptor antagonists, beta blocking agents, diuretics (ATC codes 2, 3, 7, 8, 9). Diabetes was defined as either a fasting glucose level of >7mmol/L, or self-reported or pharmacy-reported prescribed use of anti-diabetic drugs. Hypercholesterolemia was defined as a total cholesterol ≥6.21mmol/L, or self-reported use or pharmacy reported prescribed use of lipid-lowering drugs.

Statistical analysis

Statistical analyses were performed using SPSS version 22.0 (IBM corporation). Two-sided significance level was set at α=0.05.

Baseline characteristics

Baseline characteristics were compared between HRV categories using Student's t-tests, Mann-Whitney U-tests, and χ^2-tests where appropriate.

Association of HRV with CKD incidence

For this analysis participants with CKD (N=939) or unknown CKD status at baseline (N=269) were excluded. Participants were censored at death, loss to follow-up, withdrawal, or end of study. We used mid-point imputation to approximate time to event.[34] Mantel-Cox log-rank tests were performed to test for equality in hazard rates between low HRV and moderate-to-high HRV. In Cox-regression models, we adjusted for potential confounders by introducing blocks of covariates. Block 1 included age; block 2 in addition included sex, BMI, WHR, mean IBI, smoking, baseline eGFR, and baseline UAE; block 3 additionally included history of CVD, diabetes, hypertension, and hypercholesterolemia. All covariates were retained in the model; no criteria for covariate exclusion were applied.

Association of HRV with baseline levels and change in eGFR and UAE

To examine the association of baseline HRV with eGFR and UAE over time, we conducted multivariable linear mixed-effects (LME)analyses in the entire sample (N=4,605). eGFR and the natural logarithm of UAE were modelled as a function of time. Based on model fit criteria and likelihood ratio tests, we specified a base model with unstructured covariance structure, random intercept, and random slope for time.

HRV category (Q1 vs Q2-4) was added to the model to assess its association with baseline eGFR and UAE. A two-way interaction between HRV and time was introduced to assess the association of HRV with change in eGFR (ml/min/1.73m^2 per year) and

UAE (mg/24h per year). In multivariable models, we adjusted for incremental blocks of covariates as described above.

Sensitivity analyses

By design, participants with a moderately elevated urinary albumin concentration (>10mg/L) are overrepresented in the PREVEND study. To address this imbalance, we performed sensitivity analyses using statistical weights that were based on the selection probability. Also, we performed 40 imputations using the fully conditional specification method[35,36], by which we imputed missing HRV and covariate data. Additional analyses included definitions of new-onset CKD based on either impaired eGFR only (CKD_{eGFR}: eGFR<60ml/min/1.73m²) or elevated UAE only (CKD_{UAE}: UAE≥30mg/24h). Furthermore, we applied a stricter definition of the high risk group by assigning to it participants that were in Q1 of each of the three main HRV parameters, SDNN, rMSSD, and HF ("Composite low HRV", see Figure S1, Supplemental Digital Content 1). Finally, we conducted analyses on continuous measures of HRV. For these analyses all HRV parameters were transformed by their natural logarithm, which improved linearity of the associations.

RESULTS

Baseline characteristics

Baseline characteristics of the 4605 participants are presented in **Table 1**, stratified according to low vs moderate-to-high HRV (Q1 vs Q2-4), for SDNN, rMSSD, and HF (for LF, LF/HF-ratio see Table S1a, SDC 1). The medians (IQR) of the different HRV parameters are listed in **Table 2**. In univariable analyses, participants in Q1 of SDNN had lower eGFR, higher UAE at baseline, and were more likely to have CKD at baseline. Those with baseline CKD had mildly diminished eGFR (mean(sd)=81(22); eGFR<60 in 20%) and elevated UAE (mean[IQR]=43[24-89]; UAE≥30 in 70%; see **Table S2**, SDC 1). In Q1 of SDNN we observed a less favorable cardiovascular risk profile compared to Q2-4, i.e. higher prevalence of diabetes, hypertension, hypercholesterolemia, current smoking, and history of CVD. Similar results were found for other HRV measures.

In univariable comparisons between the 4605 included participants and the 2289 excluded participants of whom no valid HRV measurements were available, no relevant differences were observed in covariates or outcomes (data not shown).

Table 1. Baseline characteristics by heart rate variability categories (Q1 vs Q2-4) for the entire sample.

		SDNN			rMSSD			HF		
		Q1	Q2-4		Q1	Q2-4		Q1	Q2-4	
	Total	4.6-23 ms	23-262 ms	p	6.4-17 ms	17-377 ms	p	3.9-94 ms²	>94ms²	p
N	4605	1151	3454	n/a	1151	3454	n/a	1151	3454	n/a
Age, years	53 [45-63]	61 [53-70]	50 [43-59]	**<0.001***	60 [52-69]	51 [43-60]	**<0.001***	60 [53-69]	51 [43-59]	**<0.001***
Males, n	2270 (49%)	592 (51)%	1678 (49%)	0.094	527 (46%)	1808 (52%)	**<0.001***	519 (45%)	1816 (53%)	**<0.001***
Black race, n	28 (0.6%)	9 (0.8%)	19 (0.6%)	0.38	9 (0.8%)	19 (0.6%)	0.38	8 (0.7%)	20 (0.6%)	0.66
Height, cm	173 (9.5)	171 (9.6)	173 (9.4)	**<0.001***	172 (9.3)	173 (9.6)	**<0.001***	172 (9.4)	173 (9.5)	**<0.001***
BMI, kg/m²	26.8 (4.4)	28 (4.7)	26 (4.2)	**<0.001***	27 (4.5)	27 (4.3)	**<0.001***	27 (4.5)	27 (4.3)	**<0.001***
WHR	0.90 (0.085)	0.92 (0.081)	0.89 (0.085)	**<0.001***	0.92 (0.082)	0.90 (0.085)	**<0.001***	0.92 (0.082)	0.89 (0.084)	**<0.001***
Heart rate, beats/min	68 (10)	74 (11)	66 (8.9)	**<0.001***	75 (10)	66 (8.8)	**<0.001***	75 (10)	66 (9.0)	**<0.001***
Smoking				**<0.001***			0.28			0.23
Never, n	1315 (29%)	287 (25%)	1028 (30%)		311 (27%)	1004 (29%)		315 (28%)	1000 (29%)	
Former, n	1934 (43%)	474 (42%)	1460 (43%)		482 (43%)	1452 (43%)		478 (42%)	1456 (43%)	
Current, n	1298 (29%)	374 (33%)	924 (27%)		432 (30%)	956 (28%)		347 (30%)	951 (28%)	
SBP, mmHg	127 (19)	133 (19)	124 (18)	**<0.001***	133 (20)	124 (18)	**<0.001***	134 (19)	124 (18)	**<0.001***
DBP, mmHg	74 (9.1)	76 (8.9)	73 (9.0)	**<0.001***	77 (9.2)	72 (8.8)	**<0.001***	77 (9.0)	72 (8.8)	**<0.001***
Antihypertensive R_x, n	1019 (25%)	386 (36%)	633 (21%)	**<0.001***	335 (31%)	684 (23%)	**<0.001***	347 (32%)	672 (22%)	**<0.001***
Hypertension , n	1578 (38%)	582 (53%)	996 (33%)	**<0.001***	546 (50%)	1032 (34%)	**<0.001***	563 (52%)	1015 (33%)	**<0.001***
Fasting glucose, mmol/L	4.8 [4.4-5.3]	5.0 [4.5-5.6]	4.7 [4.4-5.2]	**<0.001***	5.0 [4.5-5.0]	4.7 [4.4-5.3]	**<0.001***	5.0 [4.5-5.5]	4.7 [4.4-5.3]	**<0.001***
Antidiabetic R_x, n	169 (4.2%)	89 (8.3%)	80 (2.7%)	**<0.001***	84 (7.9%)	85 (2.9%)	**<0.001***	86 (8.1%)	83 (2.8%)	**<0.001***
Diabetes Mellitus , n	299 (7.5%)	137 (13%)	162 (5.6%)	**<0.001***	126 (12%)	173 (5.9%)	**<0.001***	122 (12%)	177 (6.0%)	**<0.001***
History of CVD, *n*	302 (6.8%)	118 (11%)	184 (5.5%)	**<0.001***	89 (8.0%)	213 (6.4%)	0.059	100 (9.0%)	202 (6.0%)	**0.001***
Total cholesterol, mmol/L	5.5 (1.0)	5.6 (1.0)	5.4 (1.0)	**<0.001***	5.7 (1.1)	5.4 (1.0)	**<0.001***	5.7 (1.0)	5.4 (1.0)	**<0.001***
Lipid lowering R_x, n	465 (11%)	193 (18%)	272 (9.1%)	**<0.001***	158 (15%)	307 (10%)	**<0.001***	175 (17%)	290 (9.7%)	**<0.001***
Hypercholesterolemia, n	1453 (35%)	497 (45%)	956 (32%)	**<0.001***	473 (43%)	980 (32%)	**<0.001***	477 (44%)	976 (32%)	**<0.001***
Serum creatinine, mg/dL	0.82 (0.23)	0.84 (0.32)	0.82 (0.18)	0.11	0.85 (0.32)	0.81 (0.19)	**<0.001***	0.85 (0.32)	0.81 (0.19)	**<0.001***
Serum cystatin C, mg/L	0.91 (0.21)	0.99 (0.29)	0.88 (0.18)	**<0.001***	0.98 (0.28)	0.89 (0.18)	**<0.001***	0.98 (0.28)	0.89 (0.37)	**<0.001***
eGFR , ml/min/1.73m²	92 (17)*	84 (18)	94 (16)	**<0.001***	85 (18)	94 (16)	**<0.001***	85 (18)	94 (16)	**<0.001***
UAE, mg/24h	8.9 [6.2-17]	10 [6.8-22]	8.5 [6.0-15]	**<0.001***	10 [6.8-24]	8.5 [6.0-15]	**<0.001***	10 [6.8-24]	8.5 [6.0-15]	**<0.001***
Baseline CKD, n	939 (22%)	331 (30%)	608 (19%)	**<0.001***	336 (31%)	603 (19%)	**<0.001***	340 (31%)	599 (18%(	**<0.001***
Baseline $CKD_{eGFR<60}$, n	202 (4.7%)	97 (9.0%)	105 (3.3%)	**<0.001***	94 (8.8%)	972 (91%)	**<0.001***	100 (9.4%)	102 (3.2%)	**<0.001***
Baseline $CKD_{UAE\geq30}$, n	846 (18%)	283 (25%)	563 (16%)	**<0.001***	292 (26%)	554 (16%)	**<0.001***	294 (26%)	552 (16%)	**<0.001***

SDNN: standard deviation of all normal-normal RR-intervals; rMSSD: root mean square of successive differences of adjacent normal-to-normal RR-intervals; HF: high frequency power spectrum; BMI: body mass index; WHR: waist/hip ratio; SBP: systolic blood pressure; DBP: diastolic blood pressure; CVD: cardiovascular disease; Rx: medication use; eGFR: estimated glomerular filtration rate; UAE: urinary albumin excretion; CKD: chronic kidney disease, defined as estimated glomerular filtration rate (eGFR) < 60 mL/min/1.73m² or urinary albumin excretion (UAE)≥30 mg/24 hours. * indicates statistical significance (p<0.05)

Table 2. Distribution of HRV parameters.	
SDNN (ms)	31 [23-42]
rMSSD (ms)	24 [17-35]
HF (ms^2)	211 [94-454]
LF (ms^2)	242 [123-494]
LF/HF-ratio	1.2 [0.7-2.0]

HRV measures were non-normally distributed, hence data is presented as median [interquartile range]. SDNN: standard deviation of normal-to-normal RR-intervals; rMSSD: root mean square of successive differences; HF: high frequency power spectrum; LF: low frequency power spectrum.

Association of HRV with CKD incidence

We excluded those with CKD or unknown CKD status at baseline, leaving 3397 participants. Baseline characteristics for these 3397 participants are presented in Table S1b-c, SDC 1. Of these participants, 341 developed CKD during a median of 7.4 [IQR: 7.0–7.8] years of follow-up. At the earliest moment of identification, those with new-onset CKD had mildly diminished eGFR (mean[IQR]=79[59-94]; eGFR<60 in 20%) and elevated UAE (mean[IQR]=35[17-48], UAE≥30 in 72%, see Table S2, SDC 1). Event rates of CKD per HRV category are shown in **Table 3**.

Table 3. Chronic kidney disease incidence rates by heart rate variability categories (Q1 vs Q2-4).

	Total	SDNN		p	rMSSD		p	HF		p
		Q1	Q2-4		Q1	Q2-4		Q1	Q2-4	
N	3397	849	2548	n/a	849	2548	n/a	849	2548	n/a
Person-years, [IQR]	6.1 [4.6-7.3]	5.4 [2.1-7.3]	6.8 [3.1-7.4]	**<0.001**	5.9 [2.1-7.3]	6.8 [2.7-7.4]	**<0.001**	6.4 [2.3-7.4]	6.8 [3.0-7.4]	**<0.001**
New-onset CKD [a] /n (%)	341 (10%)	116 (14%)	225 (8.8%)	**<0.001**	107 (13%)	234 (9.2%)	**0.004**	109 (13%)	232 (9.1%)	**0.002**
New-onset CKD /1000 py	19.5	29.1	16.7	**<0.001**	25.9	17.5	**<0.001**	26.9	17.3	**<0.001**

Event rates by HRV category (low vs moderate-to-high HRV, Q1 vs Q2-4). SDNN: standard deviation of normal-to-normal RR-intervals; rMSSD: root mean square of successive differences of adjacent normal-normal RR-intervals; HF: high frequency power spectrum; IQR: interquartile range; CKD: chronic kidney disease; py: person-years. [a] Defined as estimated glomerular filtration rate (eGFR)<60 mL/min/1.73m^2 or urinary albumin excretion (UAE)≥30 mg/24 hours.

Incidence rate of CKD was significantly higher in those with low HRV (SDNN Q1 vs Q2-4: 29.1 v 16.7 cases per 1,000 person-years, Mantel-Cox log-rank test χ^2=23.9, df=1, p<0.001, similar for other HRV measures). The results of Cox-regression analyses are shown in **Table 4** (results for LF, LF/HF-ratio in Table S4a, SDC 1). Low HRV was associated with CKD incidence (SDNN Q1 vs Q2-4: unadjusted hazard ratio (HR)=1.66, 95%CI [1.30;2.12], similar for other HRV measures). After adjusting for confounders, this association was no longer significant (SDNN Q1 vs Q2-4: fully adjusted HR=1.13, 95%CI [0.86;1.48], similar for rMSSD, HF, and LF). Only for LF/HF-ratio a significant association was found, which remained after multivariable

adjustment (LF/HF-ratio Q1 vs Q2-4: fully adjusted HR=1.32 [1.01;1.71], p<0.043). Alternative definitions of new-onset CKD (incidence of either impaired eGFR, or of elevated UAE) yielded similar results (see **Table 4**).

Table 4. Association of heart rate variability measures (Q1 vs Q2-4) with incident chronic kidney disease.

CKD	SDNN Q1	p	rMSSD Q1	p	HF Q1	p
Unadjusted HR [95%CI]	1.66 [1.30 ; 2.12]	**<0.001***	1.51 [1.18 ; 1.93]	**0.001***	1.54 [1.20 ; 1.97]	**<0.001***
Adjusted HR [95%CI] [1]	1.02 [0.79 ; 1.32]	0.88	1.01 [0.78 ; 1.30]	0.97	0.99 [0.77 ; 1.28]	0.93
Adjusted HR [95%CI] [2]	1.10 [0.83 ; 1.45]	0.50	1.09 [0.82 ; 1.45]	0.57	1.04 [0.78 ; 1.37]	0.80
Fully adjusted HR [95%CI] [3]	1.13 [0.86 ; 1.48]	0.40	1.09 [0.82 ; 1.45]	0.55	1.02 [0.77 ; 1.35]	0.87
$CKD_{eGFR<60}$						
Unadjusted HR [95%CI]	2.44 [1.64 ; 3.63]	**<0.001***	1.92 [1.28 ; 2.88]	**0.002***	2.05 [1.37 ; 3.07]	**<0.001***
Adjusted HR [95%CI] [1]	1.05 [0.70 ; 1.59]	0.80	0.97 [0.64 ; 1.46]	0.88	0.97 [0.64 ; 1.46]	0.88
Adjusted HR [95%CI] [2]	0.90 [0.57 ; 1.42]	0.66	1.09 [0.68 ; 1.75]	0.71	0.83 [0.52 ; 1.32]	0.83
Fully adjusted HR [95%CI] [3]	0.93 [0.59 ; 1.46]	0.76	1.16 [0.72 ; 1.85]	0.54	0.89 [0.56 ; 1.41]	0.61
$CKD_{UAE\geq30}$						
Unadjusted HR [95%CI]	1.46 [1.09 ; 1.96]	**0.011***	1.43 [1.07 ; 1.92]	**0.016***	1.39 [1.04 ; 1.87]	**0.028***
Adjusted HR [95%CI] [1]	1.04 [0.76 ; 1.41]	0.82	1.07 [0.79 ; 1.45]	0.64	1.01 [0.75 ; 1.38]	0.93
Adjusted HR [95%CI] [2]	1.15 [0.83 ; 1.60]	0.40	1.23 [0.87 ; 1.73]	0.24	1.12 [0.80 ; 1.57]	0.51
Fully adjusted HR [95%CI] [3]	1.17 [0.84 ; 1.62]	0.35	1.22 [0.87 ; 1.71]	0.25	1.10 [0.79 ; 1.54]	0.56

Estimates of hazard ratios after multivariable Cox regression analysis. Reference group is moderate-to-high HRV (Q2-4). HR: hazard ratio; SDNN: standard deviation of normal-to-normal RR-intervals; rMSSD: root mean square of successive differences of adjacent normal-normal RR-intervals; HF: high frequency power spectrum; CI: confidence interval. * indicates statistical significance (p<0.05)

[1] Adjusted for age
[2] Adjusted for sex, BMI, WHR, mean IBI, smoking status, baseline eGFR, baseline UAE, in addition to above
[3] Adjusted for history of cardiovascular disease, diabetes, hypertension, and hypercholesterolemia, in addition to above.

Sensitivity analyses in imputed datasets (in which we imputed missing values of HRV and covariates), and analyses with sampling weights (to account for sampling imbalance), did not substantially change results for SDNN, rMSSD, HF, and LF (see Supplementary Tables S4b-d). However, the multivariable-adjusted HR for LF/HF-ratio was no longer significant in these analyses (LF/HF-ratio Q1 vs Q2-4: fully adjusted HR=1.19, 95%CI [0.79;1.79], in imputed datasets, similar for weighted analysis). Furthermore, a more stringent definition of the high risk group ("Composite low HRV", participants in Q1 of each of the main HRV parameters, SDNN, rMSSD, and HF, see Supplementary Table 4a) yielded similar results.

Association of HRV with baseline levels and change in eGFR and UAE

In **Table 5**, the results of LME analyses are shown for all 4605 participants (for LF and LF/HF-ratio, see Table S5a, SDC 1). Those with low HRV had significantly

Table 5. Differences between low (Q1) and moderate-to-high heart rate variability (Q2-4) measures for baseline levels and rate of decline of eGFR.

	SDNN Q1					
	Total (N=4605)	**p**	**No CKD (N=3397)**	**p**	**CKD (N=939)**	**p**
Baseline eGFR-level difference [a] (ml/min/1.73m²)						
Unadjusted β [95%CI]	-9.36 [-10.6 ; -8.08]	**<0.001***	-7.36 [-8.56 ; -6.17]	**<0.001***	-12.3 [-15.8 ; -8.74]	**<0.001***
Adjusted β [95%CI] [1]	-0.94 [-1.97 ; 0.092]	0.074	-0.60 [-1.59 ; 0.40]	0.24	-3.52 [-6.39 ; -0.66]	**0.016***
Adjusted β [95%CI] [2]	-0.81 [-1.90 ; 0.29]	0.15	-0.43 [-1.48 ; 0.63]	0.43	-4.02 [-7.05 ; -0.98]	**0.010***
Fully adjusted β [95%CI] [3]	-0.59 [-1.66 ; 0.48]	0.28	-0.42 [-1.48 ; 0.63]	0.43	-3.73 [-6.70 ; -0.75]	**0.014***
eGFR-slope difference [b] (ml/min/1.73m2 per year)						
Unadjusted β_{slope} [95%CI]	-0.068 [-0.18 ; 0.039]	0.21	-0.048 [-0.16 ; 0.063]	0.40	0.080 [-0.22 ; 0.38]	0.60
Adjusted β_{slope} [95%CI] [1]	-0.076 [-0.18 ; 0.031]	0.16	-0.061 [-0.17 ; 0.050]	0.28	0.075 [-0.22 ; 0.37]	0.62
Adjusted β_{slope} [95%CI] [2]	-0.072 [-0.18 ; 0.034]	0.18	-0.058 [-0.17 ; 0.053]	0.30	0.078 [-0.22 ; 0.37]	0.60
Fully adjusted β_{slope} [95%CI] [3]	-0.077 [-0.18 ; 0.029]	0.16	-0.059 [-0.17 ; 0.052]	0.30	0.086 [-0.21 ; 0.38]	0.57
	rMSSD Q1					
	Total (N=4605)	**p**	**No CKD (N=3397)**	**p**	**CKD (N=939)**	**p**
Baseline eGFR-level difference [a] (ml/min/1.73m²)						
Unadjusted β [95%CI]	-8.11 [-9.40 ; -6.82]	**<0.001***	-6.26 [-7.46 ; -5.05]	**<0.001***	-7.64 [-11.3 ; -3.98]	**<0.001***
Adjusted β [95%CI] [1]	-0.70 [-1.72 ; 0.32]	0.18	-0.51 [-1.48 ; 0.47]	0.31	-0.98 [-3.83 ; 1.87]	0.50
Adjusted β [95%CI] [2]	-0.90 [-2.02 ; 0.22]	0.11	-0.79 [-1.87 ; 0.29]	0.15	-1.42 [-4.56 ; 1.71]	0.37
Fully adjusted β [95%CI] [3]	-0.68 [-1.77 ; 0.42]	0.23	-0.83 [-1.91 ; 0.25]	0.13	-1.37 [-4.43 ; 1.69]	0.38
eGFR-slope difference [b] (ml/min/1.73m² per year)						
Unadjusted β_{slope} [95%CI]	-0.064 [-0.17 ; 0.043]	0.24	-0.055 [-0.17 ; 0.056]	0.33	0.22 [-0.080 ; 0.51]	0.15
Adjusted β_{slope} [95%CI] [1]	-0.068 [-0.17 ; 0.038]	0.21	-0.062 [-0.17 ; 0.048]	0.27	0.22 [-0.075 ; 0.51]	0.14
Adjusted β_{slope} [95%CI] [2]	-0.062 [-0.17 ; 0.044]	0.25	-0.059 [-0.17 ; 0.051]	0.29	0.22 [-0.075 ; 0.51]	0.15
Fully adjusted β_{slope} [95%CI] [3]	-0.064 [-0.17 ; 0.042]	0.24	-0.059 [-0.17 ; 0.051]	0.29	0.22 [-0.071 ; 0.51]	0.14
	HF Q1					
	Total (N=4605)	**p**	**No CKD (N=3397)**	**p**	**CKD (N=939)**	**p**
Baseline eGFR-level difference [a] (ml/min/1.73m²)						
Unadjusted β [95%CI]	-8.89 [-10.2 ; -7.60]	**<0.001***	-6.97 [-8.17 ; -5.77]	**<0.001***	-8.94 [-12.6 ; -5.29]	**<0.001***
Adjusted β [95%CI] [1]	-0.94 [-1.97 ; 0.085]	0.072	-0.66 [-1.64 ; 0.32]	0.19	-1.52 [-4.38 ; 1.35]	0.30
Adjusted β [95%CI] [2]	-1.11 [-2.22 ; 0.0022]	0.050	-0.82 [-1.88 ; 0.24]	0.13	-1.88 [-4.96 ; 1.20]	0.23
Fully adjusted β [95%CI] [3]	-0.76 [-1.84 ; 0.32]	0.17	-0.79 [-1.85 ; 0.27]	0.14	1.62 [-4.62 ; 1.39]	0.17
eGFR-slope difference [b] (ml/min/1.73m² per year)						
Unadjusted β_{slope} [95%CI]	-0.087 [-0.20 ; 0.021]	0.12	-0.065 [-0.18 ; 0.046]	0.25	0.21 [-0.093 ; 0.50]	0.18
Adjusted β_{slope} [95%CI] [1]	-0.090 [-0.20 ; 0.017]	0.10	-0.077 [-0.19 ; 0.034]	0.17	0.21 [-0.087 ; 0.50]	0.17
Adjusted β_{slope} [95%CI] [2]	-0.082 [-0.19 ; 0.025]	0.13	-0.075 [-0.19 ; 0.036]	0.18	0.21 [-0.089 ; 0.50]	0.17
Fully adjusted β_{slope} [95%CI] [3]	-0.087 [-0.19 ; 0.020]	0.11	-0.076 [-0.19 ; 0.035]	0.18	0.21 [-0.087 ; 0.50]	0.17

Estimates of the association between low HRV and eGFR in the total PREVEND population, and stratified for CKD at baseline, from multivariable linear mixed effects analysis. Reference group is moderate-to-high HRV (Q2-4), [a] eGFR-level: difference in baseline levels of eGFR, expressed in ml/min/1.73m², compared to reference, [b] eGFR-slope: difference in change in eGFR over time, in ml/min/1.73m² per year, compared to reference. HRV: heart rate variability; eGFR: estimated glomerular filtration rate; SDNN: standard deviation of normal-to-normal RR-intervals; rMSSD: root mean square of successive differences of adjacent normal-normal RR-intervals; HF: high frequency power spectrum; CI: confidence interval.

[1] Adjusted for age
[2] Adjusted for sex, BMI, WHR, mean IBI, smoking status, baseline UAE, in addition to above
[3] Adjusted for history of cardiovascular disease, diabetes, hypertension, hypercholesterolemia, (and baseline chronic kidney disease status in the total cohort) in addition to above.
* indicates statistical significance (p<0.05)

lower baseline levels of eGFR in the total sample (SDNN Q1 vs. Q2-4, unadjusted $\beta_{level\ difference}$=-9.36 ml/min/1.73m², 95%CI [-10.6;-8.08], p<0.001, similar for other HRV measures). However, after multivariable adjustment, the association of

low HRV with baseline eGFR was no longer significant (SDNN Q1 vs. Q2-4, fully adjusted $\beta_{\text{level difference}}$=-0.59 ml/min/1.73m², 95%CI [-1.66;0.48], p=0.28, similar for other HRV measures). During follow-up there was no significant difference in rate of decline of eGFR between HRV categories (SDNN Q1 vs Q2-4, fully adjusted $\beta_{\text{slope difference}}$=-0.077 ml/min/1.73m² per year, 95%CI [-0.18;0.029], p=0.16, similar for other HRV measures). Similarly, we found no significant association of HRV measures with UAE levels or increase (see Table S6a-b, SDC 1).

Next, we tested for a modifying effect of baseline CKD status on both level and slope by introducing their interaction terms (CKD*HRV*time; CKD*HRV; and CKD*time, in addition to their main effects) to the model. Addition of the interaction term resulted in a significant increase in log-likelihood (χ^2=64.5, Δdf=3, $p_{\text{interaction}}$<0.001 for SDNN, similar for other HRV measures), suggesting a modifying effect of baseline CKD status on the association between HRV and eGFR. Therefore, we stratified for baseline CKD status. For participants with CKD at baseline, low SDNN was associated with lower baseline eGFR. This cross-sectional association between SDNN and baseline eGFR remained after multivariable adjustment (SDNN Q1 vs Q2-4, fully adjusted $\beta_{\text{level difference}}$=-3.73 ml/min/1.73m², 95%CI [-6.70;-0.75], p=0.014). Other HRV measures did not show an association with lower baseline eGFR in this subgroup. There were no significant associations between low HRV measures and rate of renal function decline during follow-up (SDNN Q1 vs Q2-4, fully adjusted $\beta_{\text{slope difference}}$=0.086 ml/min/1.73m² per year, 95%CI [-0.21;0.38], p=0.57, similar for other HRV measures). In **Figure 1**, we show crude and adjusted estimates of baseline eGFR level (panel A) and annual eGFR change (panel B), by SDNN category and strata according to baseline CKD status.

Sensitivity analyses in imputed datasets (see Supplementary Tables S5b-c, S6c-d) yielded similar results. Application of a stricter definition of low HRV confirmed the significant result for SDNN (see Supplementary Tables S5a, S5c). Correlations (crude and age-adjusted) of HRV measures with kidney function outcomes reflected the results of our main analyses: 1) higher HRV correlated with higher baseline eGFR, but no longer after adjustment for age and 2) HRV showed no relevant correlations with eGFR slope (see **Table 6**). Results of Cox regression of continuous HRV measures supported our conclusions for the main outcome, CKD incidence. However, the association of continuous HRV with baseline levels of eGFR in CKD patients was not significant in these sensitivity analyses (see Table S7-8, SDC 1).

Table 6. Correlations between HRV parameters and kidney function outcomes

	eGFR		eGFR slope^	
	Crude	**Age-adjusted**	**Crude**	**Age-adjusted**
lnSDNN	0.276***	0.020	0.002	0.002
lnrMSSD	0.223***	-0.002	0.001	0.001
lnHF	0.254***	0.002	0.002	0.002
lnLF	0.310***	0.040**	0.003*	0.003*
lnLF/HF-ratio	0.044**	0.042**	0.001	0.000

Pearson's r and partial (age-adjusted) correlations between kidney function (eGFR and eGFR decline) and continuous, natural log-transformed HRV parameters in the total sample. * p<0.05, ** p<0.01, *** p<0.001

^ correlations for eGFR slope are standardized β's from linear mixed effects models.

All Supplementary material can be accessed using the following link www.links.lww.com/PSYMED/A436.

DISCUSSION

In this population-based, longitudinal cohort study, we examined the relation between HRVand renal outcomes. We observed an association between low HRV and higher incidence of CKD, which did not remain significant after adjustment for known CKD risk factors such as age, diabetes mellitus, and hypertension. The association between HRV and CKD risk could for a substantial part be explained by older age of those with lower HRV. An analysis of renal function over time in the total sample revealed no evidence for steeper decline in eGFR or increase in UAE in those with low HRV. In a subgroup of participants with CKD at baseline, for SDNN and a stricter definition of low HRV, we found a significant association with lower levels of baseline eGFR, which remained after adjustment for confounders, but no association with change in eGFR. For the other HRV measures (rMSSD, HF, LF, and LF/HF-ratio), we did not find significant associations with either baseline levels of eGFR or decline in eGFR during follow-up in this subgroup. These results suggest that low HRV does not contribute to CKD or to renal function decline. However, we observed that low HRV was associated with lower renal function in those that already have CKD. This implies another relation, i.e. CKD resulting in (or at least coinciding with) reduced HRV.

To our knowledge, the only comparable population-based study of HRV and its association with renal outcomes to date was conducted by Brotman et al[25]. In a sample of 13,241 adults of the ARIC cohort they observed that low HRV preceded

CKD-related hospitalization and ESRD. In our study, we could not corroborate these findings. Several differences may explain the inconsistent results. First, the endpoints and available measurements used are different: our endpoint was new-onset CKD (based on repeated measurements of serum creatinine, serum cystatin C, and UAE at each subsequent examination), whereas in ARIC, the endpoints were CKD hospitalization and ESRD. The endpoints used in ARIC imply more advanced renal disease, and are therefore a less suitable measure of de novo, likely mild, disease. Furthermore, due to the lack of baseline albumin measurements in their study, Brotman et al. could not exclude reverse causality, i.e. renal damage leading to low HRV. Second, there is a marked difference in study sample. The ARIC sample consisted of ~25% blacks, which accounted for ~50% of incident cases. This may have limited the comparability of their results to the PREVEND study, which consisted of only 0.6% blacks. A recent meta-analysis established that blacks, compared to whites, have on average higher resting values of HRV.[37] This is counter-intuitive, as black race has been associated with a higher cardiovascular risk profile[38] and risk of ESRD[39]. The ethnic differences suggest as yet unknown race-specific disease mechanisms, and stratified analyses may be warranted. Unfortunately, Brotman et al. did not explicitly adjust for race, or report race-stratified analyses. Therefore, it is unclear whether their findings also pertain to whites separately within ARIC.

Hypertension, diabetes, and cardiovascular disorders are possibly related to HRV in a bidirectional manner[13,40]. Therefore, the inclusion of these covariates in the statistical models may have led to underestimation of the effect of HRV. However, this is unlikely to have affected conclusions with regards to our main outcome, as inclusion of age almost completely explained the association between low HRV and incident CKD.

In CKD patients, we found low SDNN, and a stricter definition of low HRV, to be independently associated with lower baseline levels of eGFR, but not with steeper decline in eGFR in this subgroup. To our knowledge, the largest prospective study of HRV and disease outcomes in participants with CKD was performed by Drawz et al.[21] In 3,245 renal patients in the Chronic Renal Insufficiency Cohort (CRIC), HRV (calculated from 10s ECGs) was not independently associated with either ESRD or 50% decline in eGFR. Although we could not assess incidence of ESRD due to low numbers in our cohort, our finding that low HRV was not associated with steeper

eGFR decline is consistent with these results. In contrast, Chandra et al. did find a significant association of 24h LF/HF-ratio with incident ESRD in CKD patients.[20] However, this study was relatively small (N=305) and was a prognostic study on incidence of ESRD, rather than an etiological one, thus did not formally correct for potential confounders.[41]

In our sample of the general population, reduced HRV did not precede CKD. In contrast, we did observe an association of low SDNN, and of a stricter definition of low HRV, with low eGFR in participants that already had CKD, implying that reduced HRV is preceded by CKD. If there is any causal relationship between the two, it is more likely to be in a reversed direction (i.e. CKD causing reduced HRV). Salman recently reviewed several proposed mechanisms through which CKD could lead to increased sympathetic tone and/or decreased parasympathetic tone. Among others, these include: impaired reflex control of autonomic activity, activation of the renin-angiotensin-aldosterone system, activation of renal afferents, and mental stress in CKD[24]. Of noted interest is the potential role of social and psychological factors in the relation between CKD and HRV: e.g. mental stressors are proposed to contribute to the CKD risk factors, hypertension and diabetes, through alterations in autonomic nervous system activity and the neuro-endocrine system[42]. However, the pathophysiology underlying this relation is incompletely understood. Future work may include further characterization of these proposed mechanisms, in studies with repeated measures of autonomic and renal function as well as psychological and behavioral measures in race-stratified high-risk populations.

Major strengths of this study include the availability of serially measured creatinine and cystatin C based eGFR and 24h UAE values, which are considered to be the best parameters to define CKD, during considerable duration of follow-up. We examined multiple measures of HRV, calculated from time-series of highly standardized beat-to-beat recordings. To our knowledge, this is only the second study in the general population to examine the association of HRV with incidence of CKD, and the first to assess its effect on change in eGFR and UAE. This study is therefore an important contribution to the literature.

There were several limitations. First, HRV was calculated from time-series of pulse wave recordings. In individuals at rest, pulse rate variability is considered an accurate estimate of heart rate variability.[43] However, due to the lack of ECG

data we could not definitively exclude cardiac arrhythmias. Second, because follow-up HRV measurements were not available, we were unable to examine the association of HRV changes over time with renal disease, or vice versa. Third, HRV was missing in ~33% of participants. In an effort to minimize any bias introduced by the missingness, we conducted sensitivity analyses in multiple imputed datasets, the results of which did not change our conclusions. Although the missingness is likely random and non-problematic (e.g. due to technical failure, subject movement leading to artefacts in the recording) we cannot definitively rule out that in some cases, missing or invalid recordings may have been caused by non-random, unobserved mechanisms (e.g. cardiac arrhythmias). Fourth, estimates of GFR are less accurate in the higher range (>60 mL/min/1.73m^2). We therefore used the CKD-EPI equation for both creatinine and cystatin C, currently the best option for population-based studies.[33] Fifth, we lacked specific information on -blocking agents. This class of antihypertensive medication potentially affects both HRV and kidney function, and may therefore have caused unobserved confounding. However, we estimate -blocker user baseline prevalence to be low in this relatively healthy sample of the general population, and do not expect our conclusions to be substantially affected.

These results challenge the notion that reduced HRV represents a causal factor in CKD. Rather, they suggest that reduced HRV may be a complication of CKD.

REFERENCES

1. Chronic Kidney Disease Prognosis Consortium, Matsushita K, van der Velde M, et al. Association of estimated glomerular filtration rate and albuminuria with all-cause and cardiovascular mortality in general population cohorts: A collaborative meta-analysis. *Lancet*. 2010;375(9731):2073-2081.

2. Levey AS, de Jong PE, Coresh J, et al. The definition, classification, and prognosis of chronic kidney disease: A KDIGO controversies conference report. *Kidney Int*. 2011;80(1):17-28.

3. Levey AS, Coresh J. Chronic kidney disease. *Lancet*. 2012;379(9811):165-180.

4. Coresh J, Byrd-Holt D, Astor BC, et al. Chronic kidney disease awareness, prevalence, and trends among U.S. adults, 1999 to 2000. *J Am Soc Nephrol*. 2005;16(1):180-188.

5. Ozyilmaz A, de Jong PE, Gansevoort RT. Screening for chronic kidney disease can be of help to prevent atherosclerotic end-organ damage. *Nephrol Dial Transplant*. 2012;27(11):4046-4052.

6. DiBona GF. Neural control of the kidney: Past, present, and future. *Hypertension*. 2003;41(3 Pt 2):621-624.

7. Joles JA, Koomans HA. Causes and consequences of increased sympathetic activity in renal disease. *Hypertension*. 2004;43(4):699-706.

8. DiBona GF. Physiology in perspective: The wisdom of the body. neural control of the kidney. *Am J Physiol Regul Integr Comp Physiol*. 2005;289(3):R633-41.

9. Akselrod S, Gordon D, Ubel FA, Shannon DC, Berger AC, Cohen RJ. Power spectrum analysis of heart rate fluctuation: A quantitative probe of beat-to-beat cardiovascular control. *Science*. 1981;213(4504):220-222.

10. 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. *Circulation*. 1996;93(5):1043-1065.

11. Routledge HC, Chowdhary S, Townend JN. Heart rate variability--a therapeutic target? *J Clin Pharm Ther*. 2002;27(2):85-92.

12. Kleiger RE, Stein PK, Bigger JT,Jr. Heart rate variability: Measurement and clinical utility. *Ann Noninvasive Electrocardiol*. 2005;10(1):88-101.

13. Thayer JF, Lane RD. The role of vagal function in the risk for cardiovascular disease and mortality. *Biol Psychol*. 2007;74(2):224-242.

14. Thayer J, Hansen A, Johnsen B. Noninvasive assessment of autonomic influences on the heart: Impedance cardiography and heart rate variability. In: Luecken L, Gallo L, eds. *Handbook of physiological research methods in health psychology.* ; 2008:183-211.

15. Thayer JF, Yamamoto SS, Brosschot JF. The relationship of autonomic imbalance, heart rate variability and cardiovascular disease risk factors. *Int J Cardiol*. 2010;141(2):122-131.

16. Hillebrand S, Gast KB, de Mutsert R, et al. Heart rate variability and first cardiovascular event in populations without known cardiovascular disease: Meta-analysis and dose–response meta-regression. *Europace*. 2013;15(5):742-749.

17. Tory K, Suveges Z, Horvath E, et al. Autonomic dysfunction in uremia assessed by heart rate variability. *Pediatr Nephrol*. 2003;18(11):1167-1171.

18. Fukuta H, Hayano J, Ishihara S, et al. Prognostic value of heart rate variability in patients with end-stage renal disease on chronic haemodialysis. *Nephrol Dial Transplant*. 2003;18(2):318-325.

19. Burger AJ, D'Elia JA, Weinrauch LA, Lerman I, Gaur A. Marked abnormalities in heart rate variability are associated with progressive deterioration of renal function in type I diabetic patients with overt nephropathy. *Int J Cardiol*. 2002;86(2-3):281-287.

20. Chandra P, Sands RL, Gillespie BW, et al. Predictors of heart rate variability and its prognostic significance in chronic kidney disease. *Nephrol Dial Transplant*. 2012;27(2):700-709.

21. Drawz PE, Babineau DC, Brecklin C, et al. Heart rate variability is a predictor of mortality in chronic kidney disease: A report from the CRIC study. *Am J Nephrol*. 2013;38(6):517-528.

22. Chandra P, Sands RL, Gillespie BW, et al. Relationship between heart rate variability and pulse wave velocity and their association with patient outcomes in chronic kidney disease. *Clin Nephrol*. 2014;81(1):9-19.

23. Oikawa K, Ishihara R, Maeda T, et al. Prognostic value of heart rate variability in patients with renal failure on hemodialysis. *Int J Cardiol*. 2009;131(3):370-377.

24. Salman IM. Cardiovascular autonomic dysfunction in chronic kidney disease: A comprehensive review. *Curr Hypertens Rep*. 2015;17(8):59-015-0571-z.

25. Brotman DJ, Bash LD, Qayyum R, et al. Heart rate variability predicts ESRD and CKD-related hospitalization. *J Am Soc Nephrol*. 2010;21(9):1560-1570.

26. Pinto-Sietsma SJ, Janssen WM, Hillege HL, Navis G, De Zeeuw D, De Jong PE. Urinary albumin excretion is associated with renal functional abnormalities in a nondiabetic population. *J Am Soc Nephrol*. 2000;11(10):1882-1888.

27. Munoz ML, van Roon A, Riese H, et al. Validity of (ultra-)short recordings for heart rate variability measurements. *PLoS ONE*. 2015;10(9):e0138921.

28. Wesseling KH. Finapres: Continuous noninvasive finger arterial pressure based on the method of peňáz. In: Meyer-Sabellek W, Anlauf M, Gotzen R, Steinfeld L, eds. *Blood pressure measurement.* 1st ed. Darmstadt: Steinkopf Verlag; 1990:161-172.

29. Mulder, L. J. M.,, Roon, A. M. van,, Schweizer,D.A.,. *CARSPAN : Cardiovascular data analysis environment : User's manual.* Groningen: Iec ProGAMMA; 1995.

30. Greaves-Lord K, Tulen J, Dietrich A, et al. Reduced autonomic flexibility as a predictor for future anxiety in girls from the general population: The TRAILS study. *Psychiatry Res*. 2010;179(2):187-193.

31. Koning SH, Gansevoort RT, Mukamal KJ, et al. Alcohol consumption is inversely associated with the risk of developing chronic kidney disease. *Kidney Int*. 2015.

32. Grubb A, Blirup-Jensen S, Lindstrom V, et al. First certified reference material for cystatin C in human serum ERM-DA471/IFCC. *Clin Chem Lab Med*. 2010;48(11):1619-1621.

33. Inker LA, Schmid CH, Tighiouart H, et al. Estimating glomerular filtration rate from serum creatinine and cystatin C. *N Engl J Med*. 2012;367(1):20-29.

34. Law CG, Brookmeyer R. Effects of mid-point imputation on the analysis of doubly censored data. *Stat Med*. 1992;11(12):1569-1578.

35. Montez-Rath ME, Winkelmayer WC, Desai M. Addressing missing data in clinical studies of kidney diseases. *Clin J Am Soc Nephrol*. 2014;9(7):1328-1335.

36. van Buuren S. Multiple imputation of discrete and continuous data by fully conditional specification. *Stat Methods Med Res*. 2007;16(3):219-242.

37. Hill LK, Hu DD, Koenig J, et al. Ethnic differences in resting heart rate variability: A systematic review and meta-analysis. *Psychosom Med*. 2015;77(1):16-25.

38. Mensah GA, Mokdad AH, Ford ES, Greenlund KJ, Croft JB. State of disparities in cardiovascular health in the united states. *Circulation*. 2005;111(10):1233-1241.

39. Saran R, Li Y, Robinson B, et al. US renal data system 2015 annual data report: Epidemiology of kidney disease in the United States. *Am J Kidney Dis*. 2016;67(3 Suppl 1):Svii, S1-305.

40. Acharya UR, Joseph KP, Kannathal N, Lim CM, Suri JS. Heart rate variability: A review. *Medical and biological engineering and computing*. 2006;44(12):1031-1051.

41. Tripepi G, Jager KJ, Dekker FW, Zoccali C. Testing for causality and prognosis: Etiological and prognostic models. *Kidney Int*. 2008;74(12):1512-1515.

42. Bruce MA, Beech BM, Sims M, et al. Social environmental stressors, psychological factors, and kidney disease. *J Investig Med*. 2009;57(4):583-589.

43. Schäfer A, Vagedes J. How accurate is pulse rate variability as an estimate of heart rate variability?: A review on studies comparing photoplethysmographic technology with an electrocardiogram. *Int J Cardiol*. 2013;166(1):15-29.

PART II

Genetics of kidney function and
the translation to clinical and research practice