

**LONGITUDINAL CHANGES AND
PROGNOSTIC SIGNIFICANCE OF
CARDIOVASCULAR AUTONOMIC
REGULATION ASSESSED BY
HEART RATE VARIABILITY AND
ANALYSIS OF NON-LINEAR HEART
RATE DYNAMICS**

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OULU 2003



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Academic Dissertation to be presented with the assent of the Faculty of Medicine, University of Oulu, for public discussion in the Auditorium 8 of Oulu University Hospital, on December 5th, 2003, at 12 noon.

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2003

Abstract

Several studies have shown that altered cardiovascular autonomic regulation is associated with hypertension, diabetes, aging, angiographic severity of coronary artery disease (CAD), and increased mortality after acute myocardial infarction (AMI). The purpose of this study was to assess the temporal changes and prognostic significance of various measures of heart rate (HR) behaviour and their possible associations to coronary risk variables, and the progression of CAD in different populations.

This study comprised five patient populations. The first consisted of 305 patients with recent coronary artery bypass graft surgery (CABG) and lipid abnormalities, the second of 109 male patients with recent CABG, the third of 53 type II diabetic patients with CAD, the fourth of 600 patients with recent AMI, and the fifth of 41 elderly subjects. HR variability and non-linear measures of HR dynamics were analysed.

Among the patients with prior CABG, a significant correlation existed between the baseline HR variability (standard deviation of N-N intervals, SDNN) and the progression of CAD ($r = 0.26$, $p < 0.001$). In the longitudinal study of patients with prior CABG, only the fractal indexes of HR dynamics, such as the power law slope (β) and the short-term fractal exponent (α_1), decreased significantly. In diabetic patients, SDNN decreased significantly ($p < 0.001$) during the three-year period. The reduction of SDNN was related to cholesterol, triglyceride, and glucose levels, and also to progression of CAD ($r = 0.36$, $p < 0.01$). In the longitudinal follow-up study of patients with recent AMI, reduced fractal indices (α_1 and β), and reduced HR turbulence predicted cardiac death when measured at the convalescent phase after AMI. Reduced β and turbulence slope predicted cardiac death when measured at 12 months after AMI. In the elderly population, β ($p < 0.001$) and α_1 ($p < 0.01$) reduced significantly. Low-frequency power spectra were the only traditional measure of HR variability that decreased significantly during the 16-year period.

HR variability is associated with many risk factors of atherosclerosis and with progression of CAD among patients with ischemic heart disease. Fractal HR dynamics are more sensitively able to detect age-related changes in cardiovascular autonomic regulation. Altered fractal HR dynamics and HR turbulence are associated with increased mortality after AMI.

Keywords: acute myocardial infarction, coronary artery disease, electrocardiogram, risk factors

You`ll never walk alone

(The theme of Liverpool F.C. since 1892)

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Abbreviations

α_1	short-term fractal exponent
α_2	long-term fractal exponent
ACE	angiotensin-converting enzyme
AMI	acute myocardial infarction
ApEn	approximate entropy
β	power law slope
BB	beta blocking
BPM	beats per minute
CAD	coronary artery disease
CABG	coronary artery bypass graft surgery
ECG	electrocardiography
EF	left ventricular ejection fraction
HDL	high-density lipoprotein
HF	high frequency
HR	heart rate
LDL	low-density lipoprotein
LF	low frequency
Ln	logarithmic transformation
NYHA	New York Heart Association
QCA	quantitative coronary angiogram
R-R interval	interval between consecutive R waves
SDNN	standard deviation of R-R intervals
VLF	very low frequency
VPB	ventricular premature beat
ULF	ultra low frequency

List of original communications

This thesis is based on the following original articles, which are referred to in the text by their Roman numerals:

- I Huikuri HV, Jokinen V, Syväne M, Nieminen MS, Airaksinen KEJ, Ikäheimo MJ, Koistinen JM, Kauma H, Kesäniemi YA, Majahalme S, Niemelä KO, Frick MH (1999) Heart rate variability and progression of coronary atherosclerosis. *Arterioscler Thromb Vasc Biol* 19: 1979–1985
- II Jokinen V, Syväne M, Mäkikallio TH, Airaksinen KEJ, Huikuri HV (2001) Temporal age-related changes in spectral, fractal and complexity characteristics of heart rate variability. *Clin Physiol* 21(3): 273–281
- III Jokinen V, Ukkola O, Airaksinen KEJ, Koistinen JM, Ikäheimo MJ, Kesäniemi YA, Huikuri HV (2003) Temporal changes in cardiovascular autonomic regulation in type II diabetic patients: association with coronary risk variables and progression of coronary artery disease. *Ann Med* 35(3): 216–223
- IV Jokinen V, Tapanainen JM, Seppänen T, Huikuri HV (2003) Temporal changes and prognostic significance of measures of heart rate dynamics after acute myocardial infarction in the beta-blocking era. *Am J Cardiol* 92: 907–912
- V Jokinen V, Sourander LB, Karanko H, Huikuri HV (2003) Temporal changes in cardiovascular autonomic regulation among elderly subjects – follow-up for sixteen years. Submitted

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1 Introduction

Previous cross-sectional studies have shown that altered cardiac autonomic regulation, as assessed by heart rate (HR) variability, is associated with common cardiovascular risk factors, such as elevated blood pressure, glucose (Akselrod *et al.* 1981, Akselrod *et al.* 1985), triglyceride, and cholesterol levels (Kupari *et al.* 1993, Huikuri *et al.* 1996, Töyry *et al.* 1997, Pikkujämsä *et al.* 1998). Several studies have shown that decreased cardiac autonomic regulation of the heart is associated with hypertension, diabetes, aging, angiographic severity of CAD, and increased mortality after acute myocardial infarction (AMI) (Kleiger *et al.* 1987, Hayano *et al.* 1990, Chakko *et al.* 1993, Cowan *et al.* 1994, Pikkujämsä *et al.* 1998, Pikkujämsä *et al.* 1999).

Traditional time and frequency domain measures of HR variability have been used as non-invasive tools for assessing the autonomic tone of the heart (Akselrod *et al.* 1981, Pagani *et al.* 1986, Kleiger *et al.* 1987). In some previous studies, however, the newer methods based on fractal and complexity scaling of R-R interval variability and heart rate turbulence have been able to detect subtle changes in heart rate dynamics that are not revealed by traditional methods. These fractal and complexity measures of R-R intervals are associated with an increased risk for ventricular arrhythmias and death after AMI (Lipsitz & Goldberger 1992, Peng *et al.* 1995, Bigger, Jr. *et al.* 1996, Goldberger 1996, Huikuri *et al.* 2000, Mäkikallio *et al.* 2001a).

There are no previous longitudinal studies on the temporal changes in the autonomic cardiovascular regulation of the heart in different patient populations. Furthermore, there is limited information on the relationship between HR variability and HR dynamics, cardiovascular risk variables, and progression of coronary artery disease. The prognostic power of different measures of HR variability and HR dynamics late after AMI has not been well documented. The purpose of this study was to assess the temporal changes and prognostic significance of various measures of HR variability and HR dynamics and the possible association between HR variability, coronary risk variables, and progression of CAD in different patient populations.

2 Review of the literature

2.1 Risk factors of coronary artery disease

The most important risk factors for CAD have been shown to be high serum LDL cholesterol, high blood pressure, and smoking (Anderson *et al.* 1987, Harris *et al.* 1988, Multiple Risk Factor Intervention Trial 1996, Stamler *et al.* 2000). In the previous studies, impaired glucose control, high serum triglyceride concentration, low serum HDL cholesterol, and obesity have also been associated with the progression of CAD (Koskinen *et al.* 1992, Stamler *et al.* 1993, Haffner *et al.* 1998, Rubins *et al.* 1999). In epidemiologic studies, the insulin resistance syndrome has been shown to be related to an increased risk of coronary artery disease and to complications of ischemic heart disease (Pyörälä *et al.* 1985, Casassus *et al.* 1992, Deprés *et al.* 1994). It is also suggested that inflammatory processes may be involved in the development of atherosclerosis and its complications (Ismail *et al.* 1999). In many previous studies, mental depression has also been established as a risk factor for coronary artery disease (Barefoot *et al.* 1996, Ford *et al.* 1998). The mean cardiovascular risk factor level has decreased markedly in Finland from 1972 to 1997 (Vartiainen *et al.* 2000). However, obesity and smoking rates have increased among Finns. The previously documented decline in cholesterol levels had leveled off (Vartiainen *et al.* 2003). Nevertheless, there are some studies showing that, in older patient populations, the common risk factors do not always explain the progression of CAD (Mattila *et al.* 1988, Krumholz *et al.* 1994).

2.2 Progression of coronary artery disease

The development of clinically significant CAD is a slow process, and the first symptoms of angina pectoris usually occur at the age of 50–60 years in people living in the Nordic countries. Atherosclerosis develops in the intimal wall of the coronary artery. The local intimal dysfunction of lipid metabolism may cause atherosclerotic plaques, which are usually localized proximally to the coronary artery. The serum cholesterol concentration

plays an important role in the development of atherosclerosis, and without cholesterol, atherosclerotic plaques do not usually develop. The progression of atherosclerosis also depends on other cardiovascular risk factors, such as smoking, glucose abnormalities, aging, and hypertension (Solberg & Strong 1983, Wexler *et al.* 1996, Grundy *et al.* 1999). Previous follow-up studies with repeated angiograms have shown that baseline coronary artery stenoses progress in 20–30% of the cases within 3–4 years (Stone *et al.* 1993, Quinn *et al.* 1994, Jost *et al.* 1994), and intensive multi-factor risk reduction tends to diminish the frequency of new coronary lesions (Quinn *et al.* 1994). In a retrospective study, myocardial infarction developed more frequently from previously non-severe lesions (Ambrose *et al.* 1988).

The role of hemodynamic factors in the localized nature of coronary artery disease, i.e., the localization of coronary stenoses in specific proximal portions of the coronary arteries around the arterial branches, has been speculated based on the earlier studies, and it has been shown that hemodynamic factors may play an important role in the progression and regression of these lesions (Filipovsky *et al.* 1992). Elevated resting HR has been shown to predict cardiovascular mortality in prospective epidemiological studies (Kannel *et al.* 1987, Gillum *et al.* 1991). Ambulatory ECG recordings have shown that the minimum HR measured during a 24-hour period is even more closely related to cardiac events than resting HR or the 24-hour average HR (Perski *et al.* 1992).

2.3 Traditional measures of heart rate variability

The changes of beat-to-beat fluctuations in sinus rhythm have been called heart rate variability. The analysis of HR variability has been used as a non-invasive tool to assess cardiovascular autonomic regulation of the heart. These measurements are easy to perform, have relatively good reproducibility, and provide prognostic information on patients with heart disease (Kleiger *et al.* 1987, Casolo *et al.* 1989, Hayano *et al.* 1990, Van Hoogenhuyze *et al.* 1991, Bigger, Jr. *et al.* 1992, Bigger, Jr. *et al.* 1996, Huikuri *et al.* 1998).

2.3.1 Time domain measures of heart rate variability

The time domain measures of HR variability are calculated by statistical analyses (means and variance) from the lengths of successive R-R intervals (Task Force 1996). The most commonly used time domain indexes are the average heart rate and the standard deviation of the average R-R intervals (SDNN) calculated over a 24-hour period. This recording period is commonly used by cardiologists to calculate HR variability. SDNN is considered to reflect both the sympathetic and the parasympathetic influence on HR variability (Bigger, Jr. *et al.* 1992). Other time domain measures of HR variability are standard deviation of the means for all of the 5-minute R-R intervals covered in the recording (SDANN), the percentage of differences between adjacent R-R intervals over 50 ms (pNN50), the square root of the mean squared differences of successive R-R

intervals (RMSSD), and the standard deviation of successive differences of R-R intervals (SDSD) (Ewing *et al.* 1984, Bigger *et al.* 1989). These measures of HR variability are considered to be reliable indices of cardiac parasympathetic activity (Kleiger *et al.* 1992). SDNN of successive R-R intervals is mostly used as a time domain measure of HR variability, because the additional information from the other time domain parameters of HR variability is relatively scant. SDNN mostly reflects the very-low-frequency fluctuation in heart rate behavior. It cannot detect subtle changes in heart rate dynamics, because the fast changes in heart rate occurring within a few seconds or minutes are lost under the majority of slower changes (Goldberger 1996, Mäkikallio *et al.* 1998, Huikuri *et al.* 2000). SDNN is probably the best known HR variability index, and in recent studies, low SDNN has shown to predict mortality in post-AMI patients (Kleiger *et al.* 1987, Bigger, Jr. *et al.* 1992, Tsuji *et al.* 1994). All time and frequency domain measures of HR variability could be affected by artefacts and ectopic beats, and these measures require data from which these artefacts and ectopic beats have been eliminated.

2.3.2 Frequency domain measures of heart rate variability

The spectral analysis of HR variability gives a possibility to study the frequency-specific fluctuations of heart rate. The heart rate signal is decomposed into its frequency components and quantified in terms of their relative intensities (power) (Sayers 1973, Akselrod *et al.* 1981). The result can be displayed with the magnitude of variability as a function of frequency (power spectrum). The power spectrum reflects the amplitude of heart rate fluctuations present at different oscillation frequencies. Methods based on Fast Fourier transformation and autoregressive model estimation are used to transform heart rate signals into the frequency domain. The power spectrum of a healthy subject can usually be divided into four major frequency bands. The limits of the spectral components usually used are: HF component 0.15–0.4 Hz, LF component 0.04–0.15 Hz, VLF component 0.003–0.04 Hz, and ULF component <0.003 Hz (Task Force 1996). Total power is represented by the total area under the power spectral curve, and the power of individual frequency bands by the area under the proportion of the curve related to each band. The power of the HF, LF, VLF, and ULF components is usually expressed in absolute units (ms^2). The ratio between the LF and HF components (LF/HF ratio) is used to reflect the sympatho-vagal balance of the heart rate fluctuation (Pagani *et al.* 1986, Montano *et al.* 1994)

2.4 Non-linear analysis of heart rate dynamics

Many recent studies have shown that non-linear phenomena are involved in the genesis of HR dynamics (Sayers 1973, Goldberger & West 1987, Goldberger 1996). It has been suggested that healthy heart beat is chaotic and shows a fractal form, which may be changed upon aging and by disease (Lipsitz & Goldberger 1992, Goldberger 1996). Many authors have shown that the traditional measures of HR variability reflecting the periodic

behavior of heart rate are unable to detect subtle changes in heart rate dynamics. Therefore, new methods based on the chaos theory and the fractal behavior of heart rate fluctuations have been developed to assess the random or chaotic behavior of heart rate dynamics. These non-linear measures of HR dynamics are even better predictors of mortality in selected patient populations (Huikuri *et al.* 1998, Mäkikallio *et al.* 1999b, Huikuri *et al.* 2000) compared to the traditional measures of HR variability.

2.4.1 Detrended fluctuation analysis

Detrended fluctuation analysis (DFA) quantifies the fractal-like correlation properties of time series data (Peng *et al.* 1995, Iyengar *et al.* 1996). It characterizes heart beat fluctuations on scales of all lengths. The most commonly used variables are short-term ($\alpha_1 < 11$ beats) and long-term ($\alpha_2 > 11$ beats) scaling exponents, which quantify self-similarity over a large range of time scales. Healthy subjects have scaling exponent values between 1.1 and 1.3, indicating fractal-like behavior.

2.4.2 Power law relationship analysis of heart rate dynamics

The power law relationship of (log) power to (log) frequency is different from the traditional measures of HR variability, because it does not reflect the magnitude of HR variability, but the distribution of power-spectral density. It is calculated from the frequency range of 10^{-4} to 10^{-2} Hz, reflecting mainly fluctuations between ULF and VLF power from the power spectra. The steeper (i.e. the more negative) the slope (β) of the power law relationship is, the greater is the relative power of ULF power compared to VLF power.

2.4.3 Approximate entropy analysis

Approximate entropy (ApEn) is a measure quantifying the regularity and complexity of time series data (Pincus 1991, Pincus & Viscarello 1992, Pincus & Goldberger 1994). Low values of ApEn indicate a more regular (less complex) signal, and high values indicate irregularity (more complex) in signal behavior.

2.4.4 Heart rate turbulence analysis

Heart rate turbulence describes fluctuations of sinus rhythm cycle length after a single ventricular premature beat (VPB). Turbulence onset is, by definition, the difference between the mean of the first two sinus R-R intervals after a VPB and the last two sinus R-R intervals before the VPB divided by the mean of the last two sinus R-R intervals before the VPB. Turbulence slope is defined as the maximum positive slope of a regression line assessed over any sequence of five subsequent sinus rhythm R-R intervals within the first 20 sinus rhythm intervals after a VPB.

2.5 Reproducibility of measures of heart rate variability and heart rate dynamics

The reproducibility of HR variability measurements obtained from 24-hour ECG recordings is good in healthy subjects and in patients with heart disease (Huikuri *et al.* 1990, Van Hoogenhuyze *et al.* 1991, Hohnloser *et al.* 1992). The observed correlation coefficients have been 0.7–0.9 (mean RR interval), 0.6–0.9 (SDNN), 0.8–0.9 (HF power), and 0.8–0.9 (LF power) (Van Hoogenhuyze *et al.* 1991, Kleiger *et al.* 1991, Hohnloser *et al.* 1992). The intraindividual coefficient of variation for SDNN was $7\pm 6\%$ and that for the average RR interval $5\pm 5\%$ (Huikuri *et al.* 1990).

The comparability of the individual values of the short-term fractal exponent α_1 between 24-hour and 10-minute RR interval data has been shown to be relatively good in healthy and post-infarction subjects (correlation coefficients 0.61–0.64, $p < 0.001$) (Perkiömäki *et al.* 2001a). The relative changes in the individual values of the short-term fractal exponent α_1 ($16\pm 22\%$) and approximate entropy ($12\pm 22\%$) were substantially smaller than the change in SDNN ($40\pm 34\%$) from the acute phase of AMI to the pre-discharge period after AMI. Non-linear measurements (i.e. α_1 and approximate entropy) of HR dynamics showed less interindividual variation than the traditional HR variability measurements (SDNN) during the acute and pre-discharge periods after AMI (Perkiömäki *et al.* 2001b).

2.6 Physiological background of heart rate variability and heart rate dynamics

Various regulatory systems have significant effects on heart rate and HR variability. It is a well known fact that respiration, baroreceptor reflexes, vasomotor control, and thermoregulatory processes cause oscillations in heart rate and HR variability. Beat-to-beat fluctuations in heart rate reflect the dynamic response of these cardiovascular regulatory systems to different physiological conditions. Heart rate is usually determined by depolarization of the sino-atrial node. It is innervated with postganglionic sympathetic and parasympathetic nerve terminals, which causes continuous changes in heart rate.

The physiological mechanisms underlying the various measures of HR variability are different. Time domain measures of HR variability correlate well with vagal activity (Eckberg 1983, Hayano *et al.* 1991). The spectral HF component is associated with respiration, and this fluctuation can be abolished by atropine administration or vagotomy, which shows that parasympathetic activity is the main contributor to the HF component (Akselrod *et al.* 1981, Akselrod *et al.* 1985, Pomeranz *et al.* 1985, Pagani *et al.* 1986, Hayano *et al.* 1991). The LF component of HR variability has been associated with both sympathetic and parasympathetic activity of the autonomous nervous system (Akselrod *et al.* 1981, Akselrod *et al.* 1985, Pomeranz *et al.* 1985). LF oscillations of heart rate are reduced in patients with high sympathetic activity, i.e. with severe heart failure (van de Borne *et al.* 1997). However, most studies fail to support the association between sympathetic activity and the LF component. The sympatho-vagal balance in HR variability analysis measured by the LF/HF ratio has been questioned (Eckberg 1997).

The background of the VLF and ULF components is not well established. Atropin may abolish almost all variation of heart rate, promoting the fact that vagal activity is also a major contributor of these components (Akselrod *et al.* 1981, Pagani *et al.* 1986, Taylor *et al.* 1998). The renin-angiotensin system and changes in thermoregulation may also affect the ULF and VLF oscillations of heart rate (Sayers 1973, Akselrod *et al.* 1981, Taylor *et al.* 1998).

The physiological background of fractal scaling properties is highly speculative. There is some evidence that increased sympathetic activation is associated with impairment of the fractal dynamics of heart rate. High norepinephrine levels have been found to be associated with depression of the fractal dynamics of the heart in patients with chronic heart failure. In a recent study, intravenous infusion of norepinephrine also decreased the short-term fractal scaling exponent (Tulppo *et al.* 2001). These findings may be due to increased overall sympathetic activation of the heart.

The physiological background of heart rate turbulence is unclear. It has been suggested that, by measurement of heart rate turbulence, direct manifestation of preserved vagal antiarrhythmic protection may be captured when responding to a potentially proarrhythmic ventricular premature beat (VPB). The absent response to VPBs with high values of turbulence onset and low values of turbulence slope might be the manifestation of lost antiarrhythmic protection of the heart (Schmidt *et al.* 1999).

2.7 Correlations between different measures of heart rate variability and heart rate dynamics

All time and frequency domain measures of HR variability correlate significantly with the mean R-R interval (Van Hoogenhuyze *et al.* 1991, Kleiger *et al.* 1991, Kupari *et al.* 1993, Bigger, Jr. *et al.* 1995). Non-linear measures of HR dynamics do not correlate significantly with the average heart rate (Mäkikallio *et al.* 1996). There is a moderate correlation between the other time and frequency domain measures (Kleiger *et al.* 1991,

Bigger, Jr. *et al.* 1995), but no or only a very weak correlation between the time and frequency domain measures of HR variability and the non-linear measures of HR dynamics (Bigger, Jr. *et al.* 1996, Huikuri *et al.* 1998).

2.8 Heart rate variability and heart rate dynamics in different populations

2.8.1 Heart rate variability and heart rate dynamics in uncomplicated CAD

Heart rate variability is reduced in patients with coronary artery disease. Reduced baroreflex sensitivity has been shown to relate to coronary artery disease (Eckberg *et al.* 1971), and reduced vagal activity, i.e. altered HR variability, has been observed in patients with CAD (Airaksinen *et al.* 1987). The circadian rhythm of HR variability has been shown to be reduced in patients with CAD (Huikuri *et al.* 1994). The reduction of LF power has also been observed to correlate with the angiographic severity of CAD (Hayano *et al.* 1990). The time and frequency domain measures of HR variability are lower in patients with chronic or subacute CAD compared to healthy subjects (Bigger, Jr. *et al.* 1995). The short-term fractal scaling exponent (α_1) has been suggested to differentiate better between patients with CAD and healthy subjects compared to the traditional measures of HR variability (Mäkikallio *et al.* 1998).

2.8.2 Heart rate variability after AMI

Reduced spectral components of HR variability have been observed in patients with recent AMI. These measures are suggested to improve late after AMI, and they have been shown to continue to associate with increased post-AMI mortality (Bigger, Jr. *et al.* 1991, Bigger, Jr. *et al.* 1993).

2.8.3 Heart rate variability and diabetes

Autonomic nervous dysfunction is a common complication of diabetic subjects, and diabetes increases the risk for cardiac mortality (Koskinen *et al.* 1992, Zuanetti *et al.* 1993, Haffner *et al.* 1998). Reduced SDNN and power spectra of HR variability have been observed in patients with insulin resistance (Pikkujämsä *et al.* 1998), and cardiac autonomic dysfunction has been associated with a high risk for cardiac mortality (Rathmann *et al.* 1993, Töyry *et al.* 1997). Increased sympathetic activation has been suggested to link insulin resistance with CAD (Reaven *et al.* 1996). This theory is based

on observations showing that hyperinsulinemia is related to elevated plasma and urinary norepinephrine levels (Troisi *et al.* 1991, Arauz-Pacheco *et al.* 1996), and that subjects with insulin resistance syndrome have increased average heart rate (Stern *et al.* 1992, Facchini *et al.* 1996). Decreased HR variability may be an early marker of developing autonomic neuropathy in diabetic subjects (Töyry *et al.* 1996, Laitinen *et al.* 1999).

2.8.4 Heart rate variability and heart rate dynamics after CABG

The spectral components of HR variability have been shown to diminish after CABG (Niemelä *et al.* 1992, Hogue, Jr. *et al.* 1994, Suda *et al.* 2001). Recovery of the power spectra of HR variability after a few months of follow-up has been observed, and exercise training has been shown to improve SDNN and baroreceptor sensitivity after CABG (Iellamo *et al.* 2000, Demirel *et al.* 2002). Decreased ApEn and a moderate increase in sympathetic tone have been observed in CABG patients before the onset of atrial fibrillation (Hogue, Jr. *et al.* 1998, Dimmer *et al.* 1998). Altered HR dynamics (mostly α_1) is associated with myocardial ischemic episodes and longer treatment in an intensive care unit in patients with recent CABG (Laitio *et al.* 2000, Laitio *et al.* 2002).

2.8.5 Heart rate variability and heart rate dynamics in aging subjects

Recent cross-sectional studies have shown that the time and frequency domain measures of HR variability are related to aging, and that HR variability is lower in elderly people compared to middle-aged or young subjects (Shannon *et al.* 1987, Hayano *et al.* 1991, Bigger, Jr. *et al.* 1995, Jensen-Urstad *et al.* 1997, Pikkujämsä *et al.* 1999). Non-linear HR dynamics also show similar age dependency (Lipsitz & Goldberger 1992, Iyengar *et al.* 1996, Mäkikallio *et al.* 1998, Peng *et al.* 2002). ULF power is the only index of HR variability without age-dependent features (Bigger, Jr. *et al.* 1996).

2.8.6 Heart rate variability in other diseases

Reduced cardiovascular autonomic regulation has also been observed in many other diseases, i.e. in patients with cardiac transplantation (Sands *et al.* 1989), chronic renal failure (Vita *et al.* 1999), many neurological illnesses (Lowensohn *et al.* 1977, Kuroiwa *et al.* 1983, Korpelainen *et al.* 1996), and alcoholism (Malpas *et al.* 1991).

2.8.7 Effects of medication on heart rate variability

Beta-blocking (BB) medication has been observed to improve HR variability (mostly vagal activity) in healthy subjects (Cook *et al.* 1991), in patients with CAD (Niemelä *et al.* 1994), and in patients with recent myocardial infarction (Molgaard *et al.* 1993). BB therapy does not affect baroreceptor sensitivity in patients with CAD (Airaksinen *et al.* 1994). Diltiazem has no effect on HR variability (Cook *et al.* 1991), while digoxin may improve HR variability in healthy subjects (Kaufman *et al.* 1993). ACE inhibitors are suggested to improve HR variability after AMI (Bonaduce *et al.* 1994). Amiodarone does not affect HR variability, but propafenone and flecainide seem to reduce it (Zuanetti *et al.* 1991).

2.9 Temporal changes in heart rate variability

There is limited information on temporal changes in HR variability. One longitudinal study on healthy elderly people indicated decreased HR variability with aging (Tasaki *et al.* 2000). Spectral measures of HR variability have been suggested to improve a long time after acute myocardial infarction and after coronary bypass surgery (Bigger, Jr. *et al.* 1993, Iellamo *et al.* 2000, Demirel *et al.* 2002).

2.10 Prognostic significance of heart rate variability and heart rate dynamics

Decreased HR variability has been used in the risk stratification of post-AMI patients. A large multicenter post-AMI study showed, in 1987, that decreased HR variability predicts mortality after AMI. The relative risk of death was over 5-fold in patients with altered HR variability (SDNN<50 ms) compared to patients with preserved HR variability (SDNN>100 ms) (Kleiger *et al.* 1987). This finding has been confirmed by several other studies (Bigger, Jr. *et al.* 1992, Farrell *et al.* 1992, La Rovere *et al.* 1998). In these previous studies, HR variability indexes were measured in the post-AMI convalescent phase. It was also suggested that HR variability measured late after AMI predicts all-cause mortality (Bigger, Jr. *et al.* 1993). In selected patient populations, altered short-term fractal dynamics of heart rate has been associated with a higher risk for arrhythmic events (Mäkikallio *et al.* 1999a, Huikuri *et al.* 2000, Mäkikallio *et al.* 2001a) and increased post-AMI mortality (Tapanainen *et al.* 2002). Impaired HR variability has been observed to increase cardiac morbidity, i.e. acute myocardial infarction and unstable angina pectoris, suggesting that low HR variability measured by traditional methods is related to many adverse cardiovascular events (Tsuji *et al.* 1996). Impaired HR variability has been suggested to be a better predictor of cardiac death and arrhythmic adverse events than left ventricular ejection fraction in patients with recent AMI (Odemuyiwa *et al.* 1991, Farrell *et al.* 1992). A low scaling exponent α_1 predicted death in a series of consecutive patients

with acute myocardial infarction with or without depressed left ventricular function (Tapanainen *et al.* 2002). Reduced LF power during controlled breathing predicted independently sudden death in patients with chronic heart failure (La Rovere *et al.* 2003). Altered heart rate turbulence has also been shown to be associated with increased mortality after AMI (Schmidt *et al.* 1999).

3 Purpose of the present study

The main purpose of the present study was to assess the temporal changes and prognostic power of cardiovascular autonomic regulation in different patient populations. The specific goals of the sub-studies were:

1. to test the hypothesis that elevated HR and reduced HR variability are associated with the progression of human coronary atherosclerosis in patients with recent CABG and lipid abnormalities (I);
2. to study longitudinal changes in HR variability and HR dynamics in patients with previous CABG and the effects of various baseline variables and progression of CAD on measures of HR variability (II);
3. to assess the temporal changes in various measures of HR variability and HR dynamics and the possible association between HR variability and HR dynamics, coronary risk variables, and progression of angiographic CAD in type II diabetic subjects (III);
4. to assess the prognostic significance and temporal changes of various measures of HR variability and HR dynamics in a series of consecutive patients with AMI, for whom cardiac medication had been optimized according to the contemporary guidelines (IV), and
5. to assess temporal changes in HR variability and HR dynamics in healthy elderly subjects after follow-up for sixteen years (V).

4 Patients

The study consisted of five different patient populations. The demographic data of the different patient populations (I-V) are shown in Table 1.

The first (n=305, I) and the second (n=109, II) consisted of men aged <70 years randomized in a double-blind fashion to receive either slow-release gemfibrozil or placebo in the Lopid Coronary Angiography (LOCAT) trial, who underwent ambulatory ECG recordings before the baseline angiographic examination. The following inclusion criteria were applied: all patients had previously undergone coronary bypass surgery, and they also fulfilled the following inclusion criteria at two consecutive screening visits: HDL cholesterol ≤ 1.1 mmol/L, LDL cholesterol ≤ 4.5 mmol/L, and serum triglycerides ≤ 4.0 mmol/L. In addition, they had blood pressure $\leq 160/95$ mm Hg, body mass index ≤ 30 kg/m², left ventricular ejection fraction $\geq 35\%$, no history of diabetes, fasting glucose concentration <7.8 mmol/L, and no condition requiring therapy with calcium channel blockers, ACE inhibitors, or diuretics. All patients underwent comprehensive clinical examinations and bicycle exercise tests and received detailed dietary counseling at the baseline. Fasting serum triglycerides, cholesterol, HDL and LDL cholesterol, and blood glucose were measured at baseline and during the three-year follow-up.

The third study group (III) consisted initially of 76 eligible patients aged 42 to 65 years (mean 57 ± 6 years), who were randomly assigned to receive fenofibrate or placebo in the Diabetes Atherosclerosis Intervention Study (DAIS) in Oulu University Hospital. The final study group consisted of 53 patients after exclusions. All patients underwent ambulatory ECG recordings at baseline and after 3 years' follow-up. The inclusion criteria for the study were as follows: all patients were required to have one visible lesion in their coronary arteries in the baseline angiogram. Any previous coronary intervention was to have taken place more than 6 months before randomization. The lipid entry criteria were: a total cholesterol to HDL cholesterol ratio of four or more, in addition to either a LDL cholesterol concentration of 3.5-4.5 mmol/L and a triglyceride concentration of 5.2 mmol/l or less, or a triglyceride concentration of 5.2 mmol/l or less and LDL cholesterol of 4.5 mmol/l or less. The diagnosis of diabetes was based on a fasting plasma glucose concentration of more than 7.8 mmol/L off treatment or a plasma glucose concentration

of 11.0 mmol/l or more 2 h after a 75 g oral glucose load. The patients had no history of ketoacidosis, and their hemoglobin A_{1c} was under 170 percent of the laboratory's upper normal limit (Flapan *et al.* 1993).

The fourth group (IV) consisted of a series of consecutive patients with AMI in Oulu University Hospital. The diagnosis of AMI was confirmed by using the contemporary guidelines at the beginning of the study, with two of the following three parameters: (1) chest pain or dyspnea lasting for at least 30 min, (2) elevation of creatine kinase MB mass up to ≥ 2 x the upper limit of the reference value, and (3) ischemic electrocardiographic (ECG) changes on admission or any later change in ECG caused by AMI (i.e. > 1mm ST segment elevation or Q wave). The exclusion criteria were: advanced age (>75 years), unstable angina at recruitment, dementia, alcoholism, drug abuse, or any other condition that could impair the capacity for informed consent. A total of 600 consecutive patients (111 females and 489 males, mean age 62±10 years) with AMI fulfilled the inclusion criteria.

To optimize the treatment of these post-AMI patients, aspirin (or warfarin) and beta-blocking drugs were given to all patients, while angiotensin-converting enzyme (ACE) inhibitors or angiotensin (AT) II receptor antagonists were given to the patients with a left ventricular ejection fraction below 40%, and lipid-lowering agents to the patients with a total cholesterol level above 5.0 mmol/l, whenever no contraindications for such medications existed and the patients consented to start the medication. The cholesterol concentration was measured within the first two days after the AMI, if the patient was not on lipid-lowering medication. The dose of beta-blockers was adjusted to achieve a resting heart rate of 50-60 bpm, and the dose of ACE inhibitors (or AT II blockers) was adjusted to the dose used in randomized trials, if tolerated by the patient. Standard doses of lipid-lowering agents were used. Special attention was paid to the long-term implementation of beta-blocking medication, which was not allowed to be discontinued by the patient or the primary physician without an absolute contraindication or disabling side-effects. Percutaneous coronary angioplasty or coronary artery bypass graft surgery were performed, depending on the results of the pre-discharge exercise test, symptoms, and coronary angiography according to the guidelines. Ambulatory ECG recordings were performed between days 5 and 14 after the AMI. It was repeated at 12 months after AMI.

The fifth study population (V) consisted of subjects over 65 years old living in the City of Turku, Finland. They were participating in a large survey of the health status of the elderly. A random sample was obtained from the register of the Social Insurance Institution. No exclusion criteria other than living in an institution were used. The original population consisted of 342 subjects, and during the original 10-year follow-up, 184 subjects died. The study of this original population has been published earlier (Mäkikallio *et al.* 2001). Sixty-four patients were eligible for Holter recording after 16 years of follow-up, and the recordings of 41 were technically successful. The most common reasons for technical failure were atrial fibrillation (n=9), ectopic beats, artifacts, and in two cases, a pacemaker.

All patients were required to give informed consent, and the studies were approved by the ethical committee of the local institution.

Table 1. Baseline characteristics of the patient populations (mean(SD or %)).

Variable	Patients after CABG (n=305) (I)	CABG patients in Oulu (n=109) (II)	Patients with DM (n=53) (III)	Patients after AMI (n=600) (IV)	Elderly subjects (n=41) (V)
Age (y)	59(7)	59(7)	57(6)	62(10)	68(3)
Gender (male/female)	305/0	109/0	34/19	489/111	17/24
BMI (kg/m ²)	26(2)	26(2)	28(3)	27(4)	26(4)
Hypertensives	-	-	28(53%)	252(48%)	6(14%)
Diabetics	-	-	53(100%)	96(18%)	1(2%)
Smokers	5(6%)	6(6%)	6(11%)	175(33%)	1(2%)
BB therapy (%)	76	79	47	97	8
Systolic BP (mmHg)	136(7)	137(20)	154(21)	123(19)	156(20)
Diastolic BP (mmHg)	83(8)	83(9)	85(11)	80(11)	87(12)
Cholesterol (mmol/L)	5.2(0.7)	5.1(0.8)	5.5(0.8)	5.3(2.9)	6.7(1.4)
HDL cholesterol (mmol/L)	0.8(0.2)	0.8(0.7)	1.0(0.2)	1.1(0.3)	1.4(1.4)
LDL cholesterol (mmol/L)	3.7(0.6)	3.6(0.6)	2.8(0.6)	3.4(0.9)	4.7(1.2)
Triglycerides (mmol/L)	1.7(0.7)	1.7(0.7)	2.8(1.7)	1.6(0.9)	1.3(1.3)
Glucose (mmol/L)	4.8(0.7)	4.6(0.5)	8.5(2.8)	6.1(1.5)	5.0(1.3)

Abbreviations: BB = beta-blocking, BMI = body mass index, BP = blood pressure

5 Methods

5.1 Clinical and laboratory analysis

Clinical history, physical examination, blood pressure measurements, biochemical analyses (I-V), and exercise tests (I, II, IV) were conducted with standard methods. Serum total cholesterol, HDL and LDL cholesterol, triglyceride, and glucose were measured from overnight fasting samples (I-V) (Friedewald *et al.* 1972, R ih a *et al.* 1997, Frick *et al.* 1997, Syv anne *et al.* 1997, McGuinness *et al.* 2000).

5.2 Echocardiographic measurements

The left ventricular systolic function was measured from post-AMI patients with 2-D echocardiography at 2 to 7 days after the AMI (IV). The left ventricle was divided into 16 segments, each of which was given a score for its motion (-1 for dyskinesia, 0 for akinesia, 1 for hypokinesia, 2 for normokinesia, and 3 for hyperkinesia). Wall motion index is the mean score of all segments, and the left ventricular ejection fraction can be calculated by multiplying the wall motion index by 30 (Berning & Steensgaard-Hansen 1990).

5.3 Angiographic data

Coronary angiograms were performed at baseline and after 3 years' follow-up on patients with prior CABG and on type II diabetics (I, II, III). In patients with CABG, native coronary arteries and bypass grafts were imaged at baseline and at the end of the trial. Angiographic views and other gantry settings were recorded at baseline and reproduced exactly at follow-up. Stringent quality control of all angiography laboratories was carried out before the study and regularly during the study by an experienced third party.

Cineframes were selected for quantitative computer-assisted analysis in matching views and identical parts of the cardiac cycle, usually in end diastole or in the diastasis period, by projecting the baseline and follow-up films side by side. The images were analyzed with the Cardiovascular Measurement System (Medis) by a single trained technical analyst. The accuracy and precision of the angiographic analyses was shown to be comparable to those reported previously by other investigators (Syväne *et al.* 1994), i.e. the coefficients of variation between repeated measurements were 3.1-10.2 %. All angiographic analyses and handling of the data were done by persons blinded to the treatment group

In the diabetic study group, angiography was conducted in the Department of Cardiology in Oulu, using catheter facilities that had been initially surveyed and subsequently monitored by Medis Medical Imaging Systems B.V. (Nuenen, The Netherlands). Angiograms were done according to a standard protocol that would permit simultaneous computer-assisted quantitative analysis. Each pair of angiograms underwent rigorous quantitative analysis. The entire coronary tree was divided into segments according to the recommendations of the American Heart Association. The mean segment diameter, the minimum lumen diameter, and the mean percentage stenosis diameter were measured. Angiographic analyses were performed in the core laboratory blinded to all other data (McLaughlin *et al.* 1998).

5.4 Analysis of heart rate variability and heart rate dynamics

5.4.1 Time and frequency domain measures

Standard deviation of all R-R intervals (SDNN) from the entire recording was used as a time domain measure of HR variability. In the frequency domain analysis of HR variability, a linear detrend was applied to the R-R interval data segments of 512 samples, to make them more stationary. This was implemented by first fitting a straight line to each segment by a standard least squares method and then subtracting it from the sample value. All time and frequency domain measures of HR variability could be affected by artefacts and ectopic beats, and these measures require data from which these artefacts and ectopic beats have been eliminated. After editing of the sinus interval tachograms, the sinus interval spectrum was computed (Huikuri *et al.* 1992). A fast Fourier transform method was used to estimate the power spectrum densities of HR variability. The power spectra were quantified by measuring the areas in the following frequency bands: (1) < 0.0033 Hz (ultra-low-frequency (ULF) power) (2) 0.0033 to < 0.04 Hz (very-low-frequency (VLF) power), (3) 0.04 to 0.15 Hz (low-frequency (LF) power) and (4) 0.15 to < 0.40 Hz (high-frequency (HF) power). The ULF and VLF power spectra were analyzed and calculated from the entire recording period, while the LF and HF power spectra were analyzed from the time window of 512 R-R intervals, as recommended by the Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology (Task Force 1996). Twelve-hour (I,II) and 24-hour (III-V) average

values of LF and HF spectra were calculated from the 512 sinus interval segments. The LF/HF ratio was calculated as the ratio of LF power in ms to HF power in ms. The average HR was measured for each hour of the recording period.

5.4.2 Detrended fluctuation analysis

The detrended fluctuation analysis (DFA) technique was used to quantify the fractal-like scaling properties of the sinus interval data. The root-mean-square fluctuations of the integrated and detrended data were measured in observation windows of varying sizes and then plotted against the size of the window on a log-log scale. The scaling exponent α represents the slope of the line that relates (log) fluctuation to (log) window size. The correlation properties were measured for short-term (<11 beats, α_1) and long-term (>11 beats, α_2) fluctuations in the sinus interval data (Mäkikallio *et al.* 1998, Huikuri *et al.* 2000).

5.4.3 Power law relationship analysis

The power law relationship of R-R interval variability was calculated from the frequency range of 10^{-4} to 10^{-2} (Bigger, Jr. *et al.* 1996, Huikuri *et al.* 1998). The point power spectrum was logarithmically smoothed in the frequency domain, and the power was integrated into bins spaced $0.0167 \log$ (Hz) apart. A robust line-fitting algorithm of \log (power) on \log (frequency) was then applied to the power spectrum between 10^{-4} to 10^{-2} Hz, and the slope of this line was calculated (β).

5.4.4 Approximate entropy analysis

Approximate entropy (ApEn) is a measure quantifying the regularity or predictability of time series data. It measures the logarithmic likelihood for the runs of patterns that are close in the next incremental comparisons. A greater likelihood of remaining close produces smaller ApEn values, and conversely, random data produce higher values. Two input variables, m and r , must be fixed to compute ApEn (II,V). On the basis of the previous findings of good statistical validity, $m=2\%$ and $r=20\%$ of the standard deviation of the data sets were chosen (Pincus & Viscarello 1992, Pincus & Goldberger 1994).

5.4.5 Heart rate turbulence analysis

Heart rate turbulence refers to the fluctuations of sinus rhythm cycle length after a single ventricular premature beat (VPB). Turbulence onset is, by definition, the difference between the mean of the first two sinus R-R intervals after a VPB and the last two sinus R-R intervals before the VPB divided by the mean of the last two sinus R-R intervals before the VPB. Turbulence slope is defined as the maximum positive slope of a regression line assessed over any sequence of five subsequent sinus rhythm R-R intervals within the first 20 sinus rhythm intervals after a VPB. Both measurements were calculated for each VPB and then averaged to obtain the value for each patient (Schmidt *et al.* 1999).

5.5 Statistics

The data were analyzed using the SPSS for Windows SPSS versions 9.0 (I, II, III) and 10.1 (IV,V) (SPSS Inc., Chicago, Illinois, USA). The results were given as means (standard deviation, SD (I, III, IV,V) or standard errors of means, SEM (I, II)). The changes of continuous variables during the follow-up were analyzed by paired-sample *t*-test in each group and in all patients (II,III,IV,V). When comparisons were made between the groups, independent-samples *t*-test was used (I, II, III, IV). Pearson's bivariate correlation coefficients were used to analyze the associations between HR variables, angiographic data, and other clinical variables (I, II, III, IV). Logarithmic transformation was performed on the skewed data, i.e. spectral measures of HR variability (I, II, IV, V). These logarithmic transformations of HR variables were used in statistical analysis. One-way ANOVA was used to compare the changes of angiographic data in quartiles divided by the change of SDNN (I, III). After univariate analysis, the independent correlation between the change in SDNN and angiographic data was estimated by multiple linear regression analysis, including other coronary risk variables as cofactors (I, III).

Univariate comparisons of the baseline characteristics between the subjects with overall or cardiac death and the survivors were performed with the chi-square test for categorical variables and with the independent-samples *t*-test for continuous variables (IV). The previously described cutoff points for all HR variability measurements were used. Because there are no well defined cutoff values for the indexes measured late after AMI, optimized cutoff values based on the receiver-operating characteristics curves were used in the risk stratification late after AMI (IV). Hazards ratios and 95 % confidence intervals were calculated for each categorized HR variability measure as a predictor of overall and cardiac death in the Cox regression model. To estimate the independent power of the HR variability indexes in predicting mortality, the test results were included in the Cox proportional hazards regression analyses after stratification with age, diabetes, and functional NYHA class. Kaplan-Meier estimates of the distribution of the times from baseline to cardiac death were computed (IV). The value of $p < 0.05$ was considered significant.

6 Results

6.1 Baseline characteristics of heart rate variability and heart rate dynamics in the study groups

Table 2. The baseline values of HR dynamics in all study groups (means (SD)).

Variable	Patients after CABG (n=305) (I)	CABG patients in Oulu (n=109) (II)	Patients with DM (n=53) (III)	Patients after AMI (n=600) (IV)	Elderly subjects (n=42) (V)
Average R-R interval (ms)	973(121)	1220(140)	886(132)	916(139)	858(136)
SDNN (ms)	70(21)	70(19)	113(35)*	98(31)*	142(34)**
ULF power (ln) (ms ²)	7.9(0.6)	7.9(0.7)	9.1(0.7)*	8.6(0.5)	9.2(0.6)*
VLF power (ln) (ms ²)	6.1(0.7)	6.2(0.7)	6.7(0.8)	6.5(1.0)	6.7(0.7)
LF power (ln) (ms ²)	5.9(0.7)	5.9(0.8)	6.0(1.0)	5.4(1.2)	5.9(0.8)
HF power (ln) (ms ²)	5.0(0.7)	5.0(0.6)	5.2(1.0)	4.8(1.1)	5.2(0.8)
α_1	-	1.29(0.14)	1.19(0.18)	1.04(0.30)**	1.16(0.18)
α_2	-	1.14(0.14)	1.10(0.76)	1.08(0.15)	1.12(0.09)
β	-	-1.29(0.2)	-1.32(0.2)	-1.29(0.18)	-1.31(0.19)
ApEn	-	1.00(0.19)	1.09(0.18)	-	0.95(0.21)
Turbulence slope (ms)	-	-	-	5.5(5.8)	-
Turbulence onset (%)	-	-	-	0.27(2.4)	-

* = p<0.05, ** = p<0.01

The mean values of HR dynamics in the individual studies are shown in Table 2. SDNN was significantly lower in the patients with previous CABG and the post-AMI patients than in the other groups. ULF power was higher in the diabetic patients and in the elderly subjects. Short-term scaling exponent α_1 was significantly lower in the post-AMI patients.

6.2 Heart rate variability and progression of atherosclerosis

The mean age of the 305 male patients with prior CABG was 59 ± 7 years, and the time from CABG was 23 ± 12 months. In this study, the patients were divided into tertiles according to baseline HR variability; SDNN was 74 ± 13 ms in the lowest tertile, 104 ± 7 ms in the middle tertile, and 145 ± 25 ms in the highest tertile. The blood glucose level was higher in the tertile with the lowest SDNN than in the highest tertile, but none of the other variables, e.g., age, blood pressure, lipid values, duration of coronary artery disease, medication, presence of angina pectoris, ischemia during exercise test, or left ventricular ejection fraction, differed across the tertiles.

The progression of coronary artery stenoses, assessed from the per-patient decrease in the minimum luminal diameter of all native vessels, was more marked in the patients with the lowest SDNN than in the middle and highest tertile (Figure 1). The difference in the per-patient change in minimal luminal diameter remained significant among the HR variability tertiles after adjustments for all baseline variables, including randomization to lipid-modifying therapy (ANCOVA, $F=4.7$, $p=0.01$). In the total study group, a significant correlation existed between the baseline SDNN and the change in the minimum luminal diameter of all native vessels ($r=0.26$, $p<0.001$).

The relationship between the progression of focal coronary atherosclerosis and HR variability was only observed in the patients randomized to receive placebo treatment, but no significant relationship was observed in those receiving gemfibrozil therapy. Marked progression of focal atherosclerosis was observed mainly in the patients with the lowest SDNN in the placebo group. A significant correlation was observed between the baseline SDNN and the change in the minimal luminal diameter of all native vessels ($r=0.44$, $p<0.001$) in the placebo group. In the gemfibrozil group, no correlation was observed between the SDNN and the change in the minimum luminal diameter ($r=0.08$, NS).

Minimum HR (during sleep) was also faster in the patients with marked progression of discrete stenoses than in those with minimal progression or regression, but the maximum HR (awake) did not differ between the groups. In univariate analyses, the per-patient change in the minimum luminal diameters of the stenoses in all native vessels was related to SDNN ($p<0.0001$), triglyceride level ($p=0.009$), randomization to placebo or gemfibrozil ($p=0.003$), minimum HR ($p=0.02$), and systolic and diastolic blood pressure ($p=0.02$ for both), but not to any other measured variable. In multiple regression analysis, the change in the minimum luminal diameter was best predicted by the SDNN ($\beta=0.24$, $p=0.0001$) and triglyceride levels ($\beta=-0.16$, $p=0.009$); no other variables entered the equation.

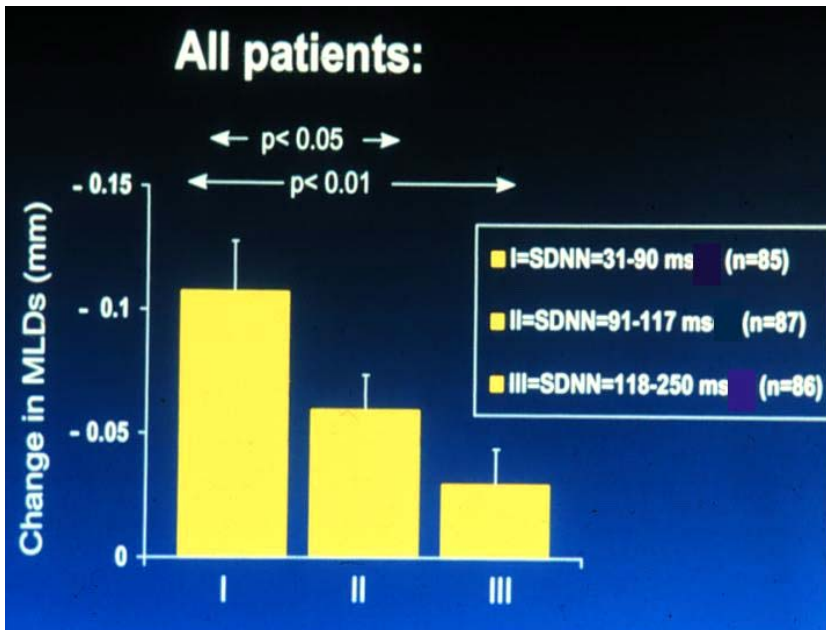


Fig. 1. Per-patient changes in the CABG study group in the minimum luminal diameter of stenoses in all native coronary arteries of the patients divided into tertiles according to the SDNN measured in 12-hour electrocardiography. Values are mean \pm SEM.

6.3 Temporal changes in heart rate variability and heart rate dynamics after CABG

Among the 109 patients with prior CABG, the traditional time and frequency domain measures of HRV did not change significantly during the 3-year follow-up. The power law slope (β) decreased from -1.29 ± 0.20 to -1.36 ± 0.23 ($p < 0.01$), and the short-term fractal exponent (α_1) of HR dynamics from 1.29 ± 0.14 to 1.22 ± 0.18 ($p < 0.001$). The approximate entropy value decreased from 1.00 ± 0.19 to 0.95 ± 0.18 ($p < 0.05$) (Table 3). The changes in HR behavior were not related to the demographic data, laboratory values, or angiographic progression of CAD. A weak correlation was observed between the change in the power law slope and the baseline glucose value ($p < 0.05$).

Table 3. The changes in HR variability and HR dynamics during the 3-year period. Means (SD).

Variable	Baseline	After 3 years
Average heart rate (bpm)	61(7)	60(7)
SDNN (ms)	70(19)	70(19)
ULF power (ms ²)	3290(2456)	4155(3501)
VLF power (ms ²)	615(387)	548(350)
LF power (ms ²)	459(346)	443(380)
HF power (ms ²)	184(147)	222(249)
α_1	1.29(0.14)	1.22(0.17)***
α_2	1.14(0.14)	1.14(0.12)
Power law slope β	-1.29(0.19)	-1.36(0.22)**
Approximate entropy	1.00(0.19)	0.95(0.18)*

* = p<0.05, ** = p<0.01, *** = p<0.001

6.4 Prognostic significance and temporal changes in heart rate variability and heart rate dynamics in type II diabetes

In type II diabetics, the 24-hour standard deviation of sinus intervals (SDNN) decreased from 113±35 ms to 94±30 ms (p<0.001) during the three-year period. The low-frequency power spectral component and the short-term fractal scaling exponent also decreased (p<0.001 and p<0.05, respectively, Table 4). The reduction of SDNN was weakly related to a change in the triglyceride level (r= -0.33, p<0.05), glucose level (r= -0.28, p<0.05), and total cholesterol concentration (r= -0.35, p<0.01). Furthermore, the reduction of SDNN was related to a decrease in the minimum lumen and mean segment diameter of the coronary arteries (r = 0.36, p<0.01, and r=0.39, p<0.01, respectively). This association was more marked in the placebo group (r=0.50, p<0.01 and r=0.44, p<0.05, respectively) than among the patients randomized to receive fenofibrate (ns for both).

Table 4. The measures of HR variability and HR dynamics in diabetic subjects at baseline and after 3 years. Means (SD).

Variable	Baseline	After 3 years
Average heart rate (bpm)	68(7)	67(6)
SDNN (ms)	113(35)	94(30)***
VLF power (ms ²)	1119(912)	837(851)
LF power (ms ²)	594(604)	372(477)***
HF power (ms ²)	272(276)	212(224)
α_1	1.19(0.18)	1.12(0.22)**

** = p<0.01, *** = p<0.001

6.5 Prognostic significance and temporal changes in heart rate variability and heart rate dynamics after AMI

During the mean follow-up period of 40 ± 12 months, 528 out of 600 post-AMI patients remained alive and 72 died (12%, annual mortality rate of 3.6 %). Death was defined as cardiac in 45 cases (7.5%). There were several differences in the clinical variables between the survivors and those who died. For example, advanced age, previously known diabetes, previous AMI, lack of thrombolytic therapy, NYHA class III or IV, and low ejection fraction were associated with cardiac death.

In univariate analysis with predefined cutoff values, reduced SDNN, ULF, and VLF spectral components as well as LF/HF ratio predicted subsequent cardiac death. Of the fractal and turbulence indexes, reduced power law slope β , short-term fractal α_1 , turbulence onset, and turbulence slope predicted cardiac mortality. In multivariate Cox proportional hazards analysis, only reduced fractal indices, both α_1 and β , as well as reduced turbulence onset and turbulence slope remained as significant predictors of cardiac and all-cause death (Table 5, Figure 2). None of the traditional time domain or spectral HR variability measures provided independent prognostic information on the risk of cardiac death (Table 5).

Measures of HR dynamics at baseline (5-7 days after AMI) and at 12 months after AMI are shown in Table 6. All time and frequency domain measures of HRV and turbulence onset increased significantly during the time course. However, the turbulence slope and the fractal measures remained unchanged. The average R-R interval did not change either.

In univariate analysis, reduced VLF spectral component, fractal indexes, and turbulence slope predicted subsequent cardiac death (Figure 3) when measured 12 months after AMI. SDNN predicted all-cause mortality, but not the occurrence of cardiac death. In multivariate analysis, only reduced β and turbulence slope remained as significant predictors of cardiac death. Reduced VLF spectral component and SDNN predicted all-cause mortality in multivariate analysis, while neither fractal indexes nor turbulence slope provided independent prognostic information on the risk of all-cause mortality (Table 7).

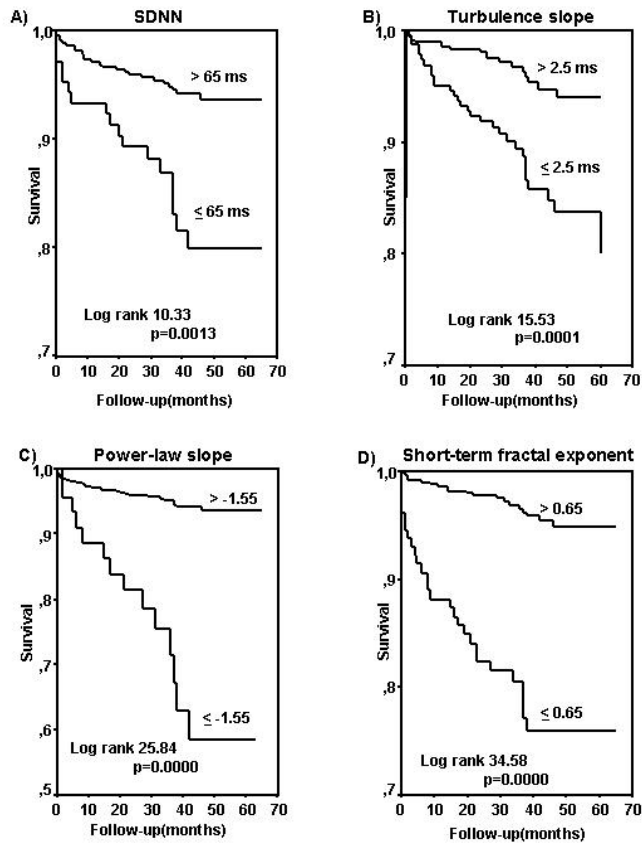


Fig. 2. Kaplan-Meier survival curves for the different parameters of HR variability and HR dynamics at the convalescent phase after acute myocardial infarction. Patients with non-cardiac death were excluded from the analysis. A. Standard deviation of N-N intervals (SDNN). B. Turbulence slope. C. Power law slope. D. Short-term fractal exponent.

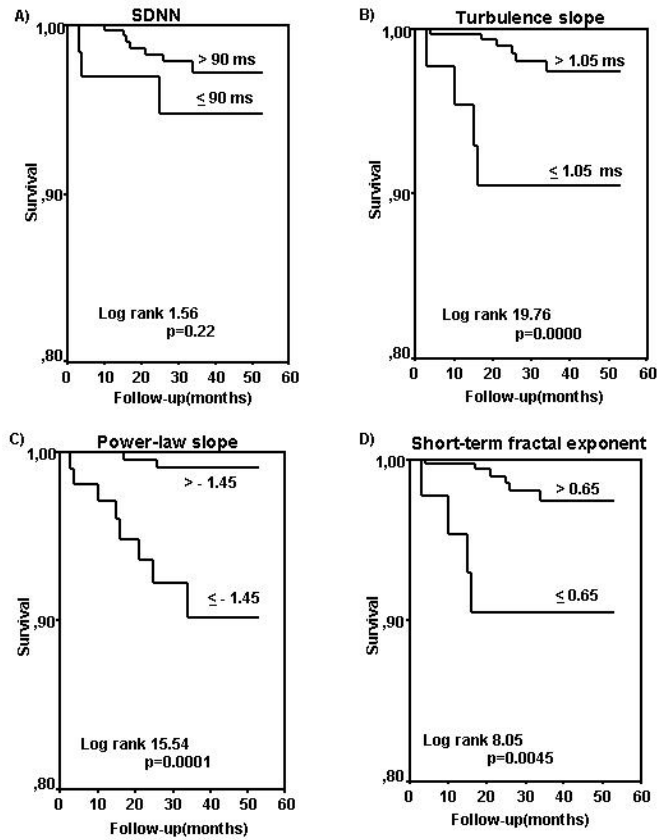


Fig. 3. Kaplan-Meier survival curves for different parameters of HR variability and HR dynamics measured at one year after acute myocardial infarction. Patients with non-cardiac death were excluded from the analysis. A. Standard deviation of N-N intervals (SDNN). B. Turbulence slope. C. Power law slope. D. Short-term fractal exponent.

Table 5. Baseline indices of HR variability and HR dynamics of post-AMI patients as predictors of subsequent mortality.

Variable	All deaths (n=72) Hazard ratio (95% CI)	Cardiac deaths (n=45) Hazard ratio (95% CI)
Univariate		
SDNN<70 (ms)	1.7(1.02-2.8)*	2.3(1.2-4.4)**
ULF ln<8.45 (ms ²)	1.9(1.1-3.0)**	2.1(1.1-3.9)*
VLF ln<5.30 (ms ²)	2.6(1.5-4.7)**	3.1(1.5-6.4)**
LF ln<3.85 (ms ²)	1.1(0.5-2.6)	1.5(0.6-3.7)
LF/HF ratio<1.45	1.8(1.1-3.0)*	2.2(1.2-4.4)*
$\alpha_1 < 0.65$	4.0(2.5-6.6)***	5.1(2.8-9.5)***
$\beta < -1.55$	4.4(2.7-7.1)***	4.3(2.3-8.0)***
Turbulence onset>0 (%)	1.8(1.1-2.9)*	2.1(1.1-4.0)*
Turbulence slope<2.5 (ms)	2.2(1.3-3.9)**	2.3(1.1-4.6)**
Multivariate		
SDNN<70 (ms)	1.1(0.7-1.9)	1.5(0.8-2.8)
ULF ln<8.45 (ms ²)	2.4(1.3-4.2)**	1.9(0.8-5.8)
VLF ln<5.30 (ms ²)	1.8(1.0-3.3)*	2.0(0.96-4.2)
LF ln<3.85 (ms ²)	1.0(0.4-2.4)	1.3(0.4-3.6)
LF/HF ratio<1.45	1.6(0.8-3.2)	1.7(0.9-3.3)
$\alpha_1 < 0.65$	2.3(1.4-3.9)**	2.7(1.4-5.2)**
$\beta < -1.55$	2.9(1.8-4.9)***	2.7(1.4-5.2)**
Turbulence onset>0 (%)	1.9(1.2-3.1)*	2.2(1.1-4.4)*
Turbulence slope<2.5 (ms)	2.2(1.3-3.9)**	2.5(1.2-5.1)*

* = p<0.05, ** = p<0.01, *** = p<0.001

Table 6. Temporal changes in the measures of HR variability and HR dynamics after acute myocardial infarction (n=416). Means (SD).

Variable	5-7 days after AMI	12 months after AMI
Average heart rate (bpm)	66(7)	64(8)
SDNN (ms)	98(31)	130(41)***
ULF power (ms ²)	7487(4810)	11642(8688)***
VLF power (ms ²)	1037(829)	1380(1105)***
LF power (ms ²)	430(447)	632(765)***
HF power (ms ²)	214(350)	333(648)**
LF/HF ratio	2.7(2.0)	2.5(1.5)
α_1	1.04(0.30)	1.04(0.26)
β	-1.29(0.18)	-1.26(0.25)
Turbulence onset (%)	0.27(2.4)	-0.92(3.4)***
Turbulence slope (ms)	5.4(5.8)	5.2(5.1)

** = p<0.01, *** = p<0.001

Table 7. HR variability and HR dynamics in post-AMI patients measured at 12 months as predictors of subsequent mortality.

Variable	All deaths (n=21) Hazard ratio (95% CI)	Cardiac deaths (n=10) Hazard ratio (95% CI)
Univariate		
SDNN<90 (ms)	5.3(1.9-14.0)***	1.9(0.2-15.2)
ULF ln<8.45 (ms ²)	2.4(0.98-6.1)	1.3(0.3-6.1)
VLF ln<6.30 (ms ²)	7.4(2.5-21.9)***	8.1(1.7-38.1)**
LF ln<3.85 (ms ²)	3.2(0.4-24.4)	3.0(0.4-23.2)
LF/HF ratio<1.45	0.7(0.1-5.1)	0.4(0.1-2.8)
α_1 <0.65	2.9(1.1-7.5)*	5.1(1.5-18.3)**
β <-1.45	2.6(1.1-6.2)*	11.6(2.5-54.8)**
Turbulence onset>0 (%)	1.0(0.4-2.3)	0.8(0.2-2.7)
Turbulence slope<1.05 (ms)	3.7(1.3-11.1)*	10.1(2.9-36.0)***
Multivariate		
SDNN<90 (ms)	4.8(1.5-15.2)*	0.9(0.1-7.8)
ULF ln<8.45 (ms ²)	2.3(0.9-5.7)	1.2(0.3-5.6)
VLF ln<6.30 (ms ²)	4.5(1.4-14.3)**	3.5(0.7-17.7)
LF ln<3.85 (ms ²)	1.5(0.2-12.1)	4.4(0.5-41.7)
LF/HF ratio<1.45)	0.6(0.2-1.5)	0.6(0.2-1.8)
α_1 <0.65	1.9(0.7-4.9)	2.9(0.8-10.5)
β <-1.45	1.7(0.7-4.1)	6.8(1.4-33.6)*
Turbulence onset>0 (%)	0.9(0.4-2.0)	0.8(0.2-2.9)
Turbulence slope<1.05 (ms)	2.9(0.9-9.0)	6.5(1.7-24.6)**

* = p<0.05, ** = p<0.01, *** = p<0.001

6.6 Temporal changes in heart rate variability and heart rate dynamics in elderly subjects

The mean age of this elderly study population was 69±4 years at the beginning of the study. Most of the subjects were female (n=27), and more than half of the subjects had some medication (n=24). In the original population of 64 patients, 9 (14%) had atrial fibrillation during the repeated Holter recording. In the final study group, 11 (14%) patients had hypertension or coronary artery disease. Body mass index (p<0.01) and cholesterol values were significantly reduced in the study group during the 16 years of follow-up (p<0.001 for both), while glucose values increased (p<0.01).

The changes that occurred in HR variability during the follow-up are shown in Table 8. SDNN did not change significantly during this period. In spectral analysis, ULF, VLF, and HF power remained unchanged during the follow-up, but LF power decreased significantly (p<0.01). In fractal analysis, the power law slope and the short-term fractal exponent α_1 decreased significantly (Figure 4), while the fractal exponent α_2 and approximate

entropy did not change markedly. The average R-R interval rose from 856 ± 137 ms to 902 ± 127 ms ($p < 0.05$).

There were no differences in HR variability between the subjects with and without cardiac medication, either at baseline or at the time of the follow-up visit. The changes in LF power, the short-term fractal exponent, and the power law slope did not correlate with each other, either. The reduction of LF power correlated strongly with the baseline glucose level ($r = -0.60$, $p < 0.001$). The reduction of the power law slope had a moderate association with higher baseline systolic ($r = 0.33$, $p < 0.05$) and diastolic blood pressure ($r = 0.44$, $p < 0.01$), i.e. the higher the blood pressure, the greater the reduction in the power law slope. The reduction of the short-term fractal exponent correlated weakly with a higher baseline body mass index ($r = 0.35$, $p < 0.05$) and a decrease of the body mass index ($r = 0.42$, $p < 0.05$). No other correlations were found between the changes in the HR variability indexes and the baseline variables.

Table 8. Temporal changes of HR variability and HR dynamics in elderly subjects after 16 years of follow-up.

Variable	Baseline	After 16 years
Average heart rate (bpm)	70(7)	67(6)*
SDNN (ms)	142(34)	133(509)
ULF power (ms^2)	11634(6010)	12504(10567)
VLF power (ms^2)	1022(701)	829(800)
LF power (ms^2)	678(654)	436(651)**
HF power (ms^2)	219(222)	268(287)
α_1	1.16(0.19)	1.06(0.18)**
α_2	1.12(0.09)	1.09(0.15)
β	-1.31(0.20)	-1.47(0.21)***
Approximate entropy	0.95(0.21)	0.95(0.18)

* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

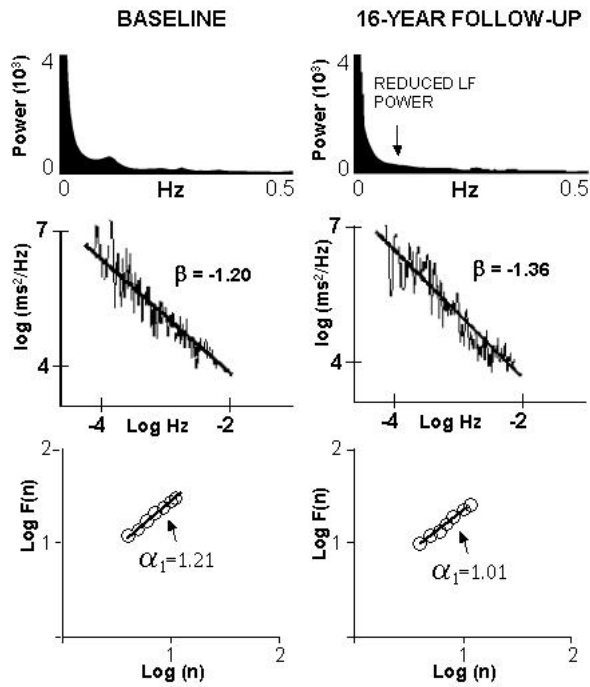


Fig. 4. An example of the power spectra of heart rate variability (upper), power law slope (β) (middle), and short-term fractal exponent (α_1) at the baseline recording (left) and 16 years after the baseline (right). LF= low-frequency.

7 Discussion

7.1 Heart rate variability and progression of atherosclerosis

Despite the epidemiological evidence of an association between low HR variability and cardiovascular mortality (Kleiger *et al.* 1987, Bigger, Jr. *et al.* 1992, Tsuji *et al.* 1996), the causes and mechanisms of this association are not well known. Follow-up and case-control studies conducted among patients after myocardial infarction have suggested that low HR variability predicts the occurrence of arrhythmic events (Hartikainen *et al.* 1996, Perkiömäki *et al.* 1997), but the results obtained in other populations suggest that reduced HR variability may also predict the occurrence of vascular events, such as angina pectoris, myocardial infarction, and coronary death (Tsuji *et al.* 1996). The present observations on patients with recent CABG provide some insight into the pathophysiology and mechanisms of the observed clinical associations, showing that reduced HR variability is related to accelerated progression of coronary atherosclerosis, rather than being a consequence of severe ischemic heart disease itself.

Elevated resting HR has been shown to predict cardiovascular mortality in a number of large-scale prospective epidemiological studies (Kannel *et al.* 1987, Gillum *et al.* 1991, Mensink & Hoffmeister 1997). Ambulatory ECG recordings have shown that the minimum HR measured during a 24-hour period is even more closely related to cardiac events than the resting HR or the 24-hour average HR (Perski *et al.* 1992), and blunted circadian rhythm of autonomic modulation of HR has been described in patients with coronary artery disease (Huikuri *et al.* 1994). In this study, the elevated minimum HR during the sleeping hours, but not the maximum HR, was found to be related to the progression of coronary artery stenoses, also providing a possible explanation for the prior epidemiological and clinical observations.

The observed associations between HR, HR variability, and the progression of focal atherosclerosis may be explained by hemodynamic factors, effects of the autonomic nervous system, or a combination of these. The role of hemodynamic factors in the localized nature of coronary artery disease, i.e., the localization of coronary stenoses to specific proximal portions of the coronary arteries around the arterial branches, has been speculated upon in the earlier studies, and it has been shown that hemodynamic factors may play

an important role in the progression and regression of these lesions (Filipovsky *et al.* 1992). The present observations support the concepts of these experimental findings by showing that reduced HR variability and elevated minimum HR predicted the progression of discrete coronary stenoses located in the proximal portions of native coronary vessels, but not the progression of diffuse disease or the development of new coronary lesions.

The autonomic nervous system may also affect coronary atherosclerosis (Kukreja *et al.* 1981, Beere *et al.* 1984). Reduced HR variability and elevated HR result from altered cardiac autonomic regulation with sympathetic predominance and/or reduced vagal tone. Increased sympathetic tone with elevated catecholamine levels may have direct effects on vascular smooth muscle cells (Yu *et al.* 1996), or it may affect other factors promoting the progression of atherosclerosis (Dzau & Sacks 1987).

The present observations fail to establish any direct causal relationship between reduced HR variability and the progression of CAD because we cannot exclude the possibility that low HR variability may be an indicator of other factors, not measured here, in relation to the progression of atherosclerosis. It is possible, for example, that there may be a genetic link between HR variability and atherogenesis, independent of hemodynamics or the autonomic nervous system.

7.2 Temporal changes in heart rate variability and heart rate dynamics after CABG

Earlier cross-sectional studies have revealed a negative correlation between age and HR variability. In healthy subjects, SDNN, VLF power, LF power, and HF power correlate inversely with aging (Bigger, Jr. *et al.* 1996, Pikkujämsä *et al.* 1999). In this study, SDNN and the spectral components of HR variability did not change in patients with recent CABG during 3 years' follow-up. One obvious reason is that the period between the recordings was relatively short. With longer follow-up, a decrease in the traditional indices of HR variability might also have occurred. Alternatively, the inverse correlation between age and the measures of HR variability may partly result from subclinical heart disease, particularly ischaemic heart disease in the elderly, which may influence the HR variability measures (Kleiger *et al.* 1987, Bigger, Jr. *et al.* 1992, Huikuri *et al.* 1994). In cross-sectional studies, where the subjects do not serve as their own controls, there may also be other baseline differences between the study groups, which may potentially influence the measures of HR variability (Lipsitz & Goldberger 1992, Umetani *et al.* 1998).

In this study, α_1 , power-law slope β , and ApEn decreased significantly during the follow-up. These measures of HR variability reflect different aspects of HR dynamics compared with the traditional measures of HR variability. They do not indicate the magnitude of HR fluctuations around its mean value, but rather the scaling characteristics and other features of the behavior. The short-term fractal exponent α_1 reflects the correlation properties of short-term R-R interval fluctuations (Ho *et al.* 1997, Mäkikallio *et al.* 1998, Huikuri *et al.* 2000, Mäkikallio *et al.* 2001a), and the power law slope β indicates the sca-

ling properties of R-R interval data over long periods (Bigger, Jr. *et al.* 1996, Huikuri *et al.* 1998). ApEn quantifies the regularity and complexity of time series (Pincus 1991, Pincus & Viscarello 1992, Pincus & Goldberger 1994).

The temporal changes in the fractal and complexity properties of HR dynamics were not related to changes in the laboratory values or progression of CAD, suggesting that these changes are related to aging itself. Only a weak correlation was found between the fasting glucose concentration and the changes in the power law slope β . Previous studies have shown the correlation between glucose tolerance and several features in HR variability (Töyry *et al.* 1997, Huikuri *et al.* 1998, Laitinen *et al.* 1999). The present data suggest that glucose metabolism may also affect the long-term fractal correlation properties of HR dynamics.

7.3 Temporal changes in heart rate variability, heart rate dynamics, and progression of CAD in type II diabetes

Overall HR variability, measured as SDNN, decreased significantly in the type II diabetic subjects even during the relatively short period of three years, suggesting rapid deterioration of cardiovascular autonomic function in type II diabetes. To our knowledge, there are no previous longitudinal studies regarding heart rate variability variables measured from 24-hour Holter recordings in patients with type II diabetes. In the spectral and fractal analysis of HR variability, the most marked changes in HR dynamics were observed in the LF frequency spectral component and the short-term fractal scaling exponent. Notably, the HF spectral component, reflecting the cardiac vagal outflow, did not decrease significantly during the 3-year period. In experimental studies, reduced short-term scaling exponent and blunted LF oscillations of HR have been shown to be closely associated with elevated levels of circulating norepinephrine and thereby possibly to reflect the increased sympathetic activity (Tulppo *et al.* 2001). These observations suggest that changes in HR variability, particularly the blunted LF oscillations of HR and the altered short-term fractal properties of HR dynamics, may reflect an increase of sympathetic activity rather than a decrease of vagal outflow in diabetic patients over time.

The reasons for the rapid reduction of overall HR variability in type II diabetes are speculative. HR variability has been shown to be associated with elevated systolic blood pressure (Huikuri *et al.* 1996), impaired glucose balance (Laitinen *et al.* 1999, Stein *et al.* 2000, Pikkujämsä *et al.* 2001), and elevated lipid values, e.g. plasma triglyceride level (Pikkujämsä *et al.* 1998). In this study, glucose control did not change significantly. Blood pressure and lipid values had a trend toward improved values. Despite this, HR variability decreased during the 3-year period, more markedly among the patients who did not show improvement in their lipid profile. In fact, SDNN also decreased in the fenofibrate group despite the marked improvement in the lipid profile. Thus, it is evident that rapid reduction of HR variability is a specific feature related to type II diabetes itself. This impairment in autonomic regulation may also be one of the factors related to the increased risk of cardiovascular events in type II diabetic subjects.

A reduction of HR variability was also associated with more rapid progression of both discrete and diffuse coronary lesions assessed by quantitative coronary angiography. This association may well be explained by the fact that the impairment of HR variability was weakly associated with several risk factors of atherosclerosis, such as cholesterol, triglyceride, and glucose levels. Insulin resistance syndrome, which itself is associated with altered autonomic regulation (Pikkujämsä *et al.* 1998), may also be a common denominator of the rapid progression of CAD and autonomic dysfunction.

7.4 Prognostic power and temporal changes in heart rate variability and heart rate dynamics after AMI

Numerous previous studies assessing the prognostic power of time domain and spectral indexes have shown that these parameters predict mortality when measured at the convalescent phase after AMI (Kleiger *et al.* 1987, La Rovere *et al.* 1998, Huikuri *et al.* 2000). Most of these studies are from the era without frequent usage of BB medication. For example, in the largest prospective study, Autonomic Tone and Reflexes After Acute Myocardial Infarction ATRAMI (La Rovere *et al.* 1998), only 20% of the patients were on BB medication. BB drugs have significant effects on various measures of HR variability as well as on post-AMI mortality (Yusuf *et al.* 1985, Cook *et al.* 1991, Niemelä *et al.* 1994, Sandrone *et al.* 1994, Airaksinen *et al.* 1996, Mortara *et al.* 2000). Therefore, the prognostic value of traditional HR variability measures in the current treatment era is not well established. Concurrently with the present findings, our previous analysis, including post-AMI patients from two different centers, showed that traditional HR variability indexes lose some of their independent predictive value in the presence of current treatment (Tapanainen *et al.* 2002).

Apart from optimized BB treatment, there are also other salient differences between the present and the previous studies. The present research consisted of a single-center study with uniform treatment of post-AMI patients and with cardiac death as the major end-point. This was a pre-defined end-point, because it may provide more relevant clinical information than analysis of all-cause mortality. Prediction of death due to all causes may have less relevance for therapeutic decisions, e.g. for designing antiarrhythmic or other preventive cardiovascular interventions for post-AMI patients. Furthermore, we used multivariate analysis in estimating the predictive power of various HR variability indexes, because it may be more informative to know which of the variables provide prognostic information, independent of clinical estimates.

In recent studies, the fractal analysis of HR variability and HR turbulence have been shown to provide more powerful prognostic information than traditional HR variability analysis (Schmidt *et al.* 1999, Huikuri *et al.* 2000, Mäkikallio *et al.* 2001b, Ghuran *et al.* 2002). These studies have mostly included patients from the pre-BB era (Schmidt *et al.* 1999, Ghuran *et al.* 2002) or from selected groups of post-AMI patients (Huikuri *et al.* 2000, Mäkikallio *et al.* 2001b). In this study, both short-term and long-term fractal scaling exponents, turbulence onset, and turbulence slope predicted overall and cardiac mortality even after adjustment for clinical variables. In contrast to the traditional HR variability

indexes, fractal indexes, turbulence onset, and turbulence slope predicted cardiac mortality, confirming that these indexes of HR dynamics retain their prognostic power in the BB era, even after adjustment for clinical variables.

Previous studies have shown that time domain and spectral measures of HR variability improve over time after AMI (Lombardi *et al.* 1987, Vaage-Nilsen *et al.* 2001). Concurrent with these observations, SDNN and spectral measures improved significantly during the one-year period in the present population. Similarly to the time and frequency domain measures of HR variability, turbulence onset improved significantly, but turbulence slope remained more stable. Fractal indexes of HR dynamics also remained unchanged.

There is less information on the predictive power of HR variability among patients with an old infarction. One previous study from the pre-thrombolytic era showed that both time-domain and spectral measures continued to predict all-cause mortality even when measured late after AMI (Bigger, Jr. *et al.* 1993). Of the traditional HR variability indexes, only the very-low-frequency spectral component provided information on the risk of cardiac death in the present study. SDNN continued to predict all-cause mortality late after AMI, but not specifically the risk for future cardiac death. Both fractal indexes and turbulence slope predicted cardiac death, and power law slope and turbulence slope remained independent predictors of cardiac mortality after adjustments for clinical variables. Neither fractal HR indexes nor turbulence slope predicted the occurrence of non-cardiac death late after AMI.

7.5 Temporal changes in heart rate variability and heart rate dynamics in elderly subjects

Several earlier studies have shown that the magnitude of total HR variability, measured by traditional methods, is lower among elderly subjects than healthy middle-aged or young subjects (Hayano *et al.* 1991, Bigger, Jr. *et al.* 1995, Pikkujämsä *et al.* 1999). These studies have included relatively small samples of elderly subjects, usually ones aged under 80 years. The present long-term longitudinal study shows that overall HR variability, measured by SDNN, does not show any further reduction after the age of 70 years.

LF power was the only spectral measure of HR variability that decreased significantly over time. The LF spectral component has been associated with both sympathetic and parasympathetic activity of the autonomic nervous system (Akselrod *et al.* 1985). LF oscillations of HR typically result from baroreflex regulation of HR in response to spontaneous fluctuation of blood pressure (Madwed *et al.* 1989). Thereby, the reduction in the LF spectral component may well result from the attenuation of the baroreflex control of HR in the elderly. Alternatively, the reduced LF spectral component may also be a result of enhanced sympathetic activity. Although the reduction of the LF spectral component is usually associated with reduced sympathetic activity, when measured under controlled conditions using passive head-up tilt (Pagani *et al.* 1986), there is increasing evidence to suggest that the reduced LF spectral component is due to sympatho-excitation, when measured from Holter recordings in free-running conditions. This is explained by the saturation of the LF oscillations of HR during the elevated sympathetic drive. This pheno-

menon has been described in, for example, heart failure patients, who have almost absent LF oscillation of HR despite high sympathetic activity (van de Borne *et al.* 1997). Reduced LF power is also associated with an increased risk of death among patients with chronic heart failure (La Rovere *et al.* 2003). Thus, the present observations of a reduction in the LF oscillations of HR may be partly explained by the increase of sympathetic activity upon aging.

Power law slope and short-term fractal exponent decreased significantly over time among the present elderly subjects. Power law slope and short-term fractal exponent differ from the traditional measures of HR variability in that they mainly reflect the distribution of the spectral characteristics of HR variability rather than the magnitude of HR variability. Short-term fractal exponent reflects the scaling characteristics of HR fluctuations over short periods, while power law slope represents the characteristics of HR dynamics over long-range time scales. According to the present findings, these scaling characteristics of HR dynamics clearly show temporal changes among the elderly despite the stable overall magnitude of HR variability. These changes in fractal-like HR dynamics may reflect some impairment in the cardiovascular regulation systems and possibly imply an increased risk of untoward cardiac events.

There were no correlations between the changes in the LF spectral component, power law slope, and short-term scaling exponent, showing that these indexes clearly describe different aspects of HR behavior. The reduction of LF power correlated strongly with elevated baseline glucose level, suggesting that the elderly subjects with impaired glucose metabolism undergo more rapid deterioration of baroreflex-mediated control of heart rate or, alternatively, more rapid increase of sympathetic drive upon aging. Diabetes itself causes altered autonomic function, and many previous studies have shown glucose metabolism to be associated with many alterations in HR variability (Töyry *et al.* 1996, Pikkujämsä *et al.* 1998, Laitinen *et al.* 1999).

Reduction of the power law slope correlated with elevated blood pressure. In previous cross-sectional studies, elevated blood pressure has been associated with decreased overall HR variability (Pikkujämsä *et al.* 1998). It has been suggested that hypertensive subjects have lower arterial compliance, which may cause alterations in cardiovascular autonomic regulation (Kingwell *et al.* 1995). The reduction of the short-term fractal exponent α_1 correlated with the reduction of BMI during the 16 years of follow-up. Reduction of weight may well be a sign of frailty and impaired overall health, possibly explaining the altered short-term dynamics of HR.

In many recent studies, altered fractal measures of HR dynamics have been shown to be associated with increased mortality in different patient populations (Huikuri *et al.* 1998, Mäkikallio *et al.* 1999, Huikuri *et al.* 2000). The loss of the fractal nature of HR dynamics predicts a poor prognosis, and the present study showed that, even among elderly subjects, the fractal characteristics of HR behavior change toward more random short-term fractal dynamics and steeper power law slope, which both are signs of a poor prognosis.

7.6 Methodological limitations

This thesis is based on observational sub-studies on different patient populations. Observational studies may not yield definite information on the cause-effect relations between various phenomena, such as low HR variability and the progression of atherosclerosis or abnormal HR dynamics and mortality observed in the present studies. Therefore, these observations should be confirmed in future randomized trials with interventions.

Computer-assisted quantitative coronary arteriography (QCA) was used in the studies I and III to assess the progression or regression of coronary atherosclerosis. This method has gained widespread acceptance in assessing changes in coronary dimensions over time. However, in a previous study, the correlation coefficient between QCA and panel-based estimates of the size of an occluded lesion in the coronary artery was 0.70 and that for a change of lesion size 0.28 (Mack *et al.* 1992). It has been suggested that changes of 0.4 millimetres or more for the minimum diameter and 15 % or more for the stenosis diameter, measured quantitatively, are recommended as criteria of the progression and regression of coronary artery disease (Waters *et al.* 1993). A change of 0.40-0.48 millimetres or more in the minimum luminal diameter represents true progression or regression of coronary atherosclerosis with more than 95 % confidence (Lesperance *et al.* 1992, Syväne *et al.* 1994). In this study, all coronary angiograms were performed by an experienced cardiologist, and the angiographic data were analyzed in the core laboratory to obtain consistent results. In study III, the mean correlation coefficients for minimum lumen diameter were 0.98 for intraobserver variability, 0.77 for inter-observer variability, and 0.96 for inter-angiogram variability. For segment length, the corresponding values were 0.99, 0.79, and 0.94 (McLaughlin *et al.* 1998).

The prognostic significance of different measures of HR variability and HR dynamics was assessed in study IV. When a diagnostic test is a continuous variable, as in this study, it is necessary to use cut-off points for prognostic evaluations. We used pre-defined cut-off points for different measures of HR variability and HR dynamics whenever they were available. The other cut-off points were measured for all-cause mortality from receiver-operating characteristics curves. The optimal cut-off points of different HR variables for mortality were low, because of the low mortality rate. In populations with low mortality, even the relatively good tests lose some of their prognostic power. However, both fractal indexes and turbulence slope predicted cardiac death at the convalescent and late phase after AMI. The sample size in study IV was small, and the results must be considered preliminary, because of the relatively low event rate late after AMI. Further follow-up studies using different measures of HR variability and HR dynamics as pre-defined risk markers will be needed to establish the clinical utility of these measurements for routine use.

8 Conclusions

This study showed that the temporal changes in HR variability and HR dynamics depend strongly on the baseline characteristics of the patients, and that altered HR variability and HR dynamics have prognostic significance for the progression of CAD and mortality after AMI. The specific findings in the substudies were as follows:

1. Low HR variability analyzed from ambulatory ECG predicts rapid progression of coronary artery disease in patients with prior CABG. HR variability provides information on the progression of focal coronary atherosclerosis beyond that obtained by traditional risk markers of atherosclerosis.
2. The fractal characteristics of HR dynamics and the complexity properties of R-R intervals undergo rapid changes along with aging, and the fractal and complexity analysis methods are more sensitive than the methods of traditional analysis in documenting temporal age-related changes in HR behavior among patients with previous CABG.
3. Cardiovascular autonomic regulation, as assessed by HR variability and HR dynamics, deteriorates rapidly in type II diabetic subjects with CAD over time. Impairment in HR variability is associated with changes in the common coronary risk variables and with progression of CAD.
4. Traditional time domain and spectral measures of HR variability and turbulence onset improved significantly during the time course after the AMI, while the fractal HR dynamics and turbulence slope remained stable. Fractal HR variability and HR turbulence retained their prognostic power in the BB era, when measured either at the convalescent or the late phase after the AMI.
5. The magnitude of total HR variability and the respiratory vagal modulation of HR do not change over time in the elderly, but the low-frequency oscillations of HR and fractal HR behavior undergo alterations.

References

- Airaksinen KE, Ikäheimo MJ, Linnaluoto MK, Niemelä M & Takkunen JT (1987) Impaired vagal heart rate control in coronary artery disease. *Br Heart J* 58: 592–597.
- Airaksinen KE, Ikäheimo MJ, Niemelä MJ, Valkama JO, Peuhkurinen KJ & Huikuri HV (1996) Effect of beta blockade on heart rate variability during vessel occlusion at the time of coronary angioplasty. *Am J Cardiol* 77: 20–24.
- Airaksinen KE, Niemelä MJ & Huikuri HV (1994) Effect of beta-blockade on baroreflex sensitivity and cardiovascular autonomic function tests in patients with coronary artery disease. *Eur Heart J* 15: 1482–1485.
- Akselrod S, Gordon D, Madwed JB, Snidman NC, Shannon DC & Cohen RJ (1985) Hemodynamic regulation: investigation by spectral analysis. *Am J Physiol* 249: H867–H875.
- Akselrod S, Gordon D, Ubel FA, Shannon DC, Berger AC & Cohen RJ (1981) Power spectrum analysis of heart rate fluctuation: a quantitative probe of beat-to-beat cardiovascular control. *Science* 213: 220–222.
- Ambrose JA, Tannebaum MA, Alexopoulos D, Hjemdahl-Monsen O, Leavy J, Weiss M, Borricco S, Gorlin R & Fuster V (1988) Angiographic progression of coronary artery disease and the development of myocardial infarction. *J Am Coll Cardiol* 12: 56–62.
- Anderson KM, Castelli WP & Levy D (1987) Cholesterol and mortality. 30 years of follow-up from the Framingham study. *JAMA* 257: 2176–2180.
- Arauz-Pacheco C, Lender D, Snell PG, Huet B, Ramirez LC, Breen L, Mora P & Raskin P (1996) Relationship between insulin sensitivity, hyperinsulinemia, and insulin-mediated sympathetic activation in normotensive and hypertensive subjects. *Am J Hypertens* 9: 1172–1178.
- Barefoot JC & Schroll M (1996) Symptoms of depression, acute myocardial infarction, and to mortality in a community sample. *Circulation* 93: 1976–1980.
- Beere PA, Glagov S & Zarins CK (1984) Retarding effect of lowered heart rate on coronary atherosclerosis. *Science* 226: 180–182.
- Berning J & Steensgaard-Hansen F (1990) Early estimation of risk by echocardiographic determination of wall motion index in an unselected population with acute myocardial infarction. *Am J Cardiol* 65: 567–576.
- Bigger JT jr., Albrecht P, Steinman RC, Rolnitzky LM, Fleiss JL & Cohem RJ (1989) Comparison of time- and frequency domain-based measures of cardiac parasympathetic activity in holter recordings after myocardial infarction. *Am J Cardiol* 64: 536–538.
- Bigger JT, Jr., Fleiss JL, Rolnitzky LM & Steinman RC (1993) Frequency domain measures of heart period variability to assess risk late after myocardial infarction. *J Am Coll Cardiol* 21: 729–736.

- Bigger JT, Jr., Fleiss JL, Rolnitzky LM, Steinman RC & Schneider WJ (1991) Time course of recovery of heart period variability after myocardial infarction. *J Am Coll Cardiol* 18: 1643–1649.
- Bigger JT, Jr., Fleiss JL, Steinman RC, Rolnitzky LM, Kleiger RE & Rottman JN (1992) Frequency domain measures of heart period variability and mortality after myocardial infarction. *Circulation* 85: 164–171.
- Bigger JT, Jr., Fleiss JL, Steinman RC, Rolnitzky LM, Schneider WJ & Stein PK (1995) RR variability in healthy, middle-aged persons compared with patients with chronic coronary heart disease or recent acute myocardial infarction. *Circulation* 91: 1936–1943.
- Bigger JT, Jr., Steinman RC, Rolnitzky LM, Fleiss JL, Albrecht P & Cohen RJ (1996) Power law behavior of RR-interval variability in healthy middle-aged persons, patients with recent acute myocardial infarction, and patients with heart transplants. *Circulation* 93: 2142–2151.
- Bonaduce D, Marciano F, Petretta M, Migaux ML, Morgano G, Bianchi V, Salemme L, Valva G & Condorelli M (1994) Effects of converting enzyme inhibition on heart period variability in patients with acute myocardial infarction. *Circulation* 90: 108–113.
- Casassus P, Fontbonne A, Thibault N, Ducimetiere P, Richard JL, Claude JR, Warnet JM, Rosselin G & Eschwege E (1992) Upper-body fat distribution: a hyperinsulinemia-independent predictor of coronary heart disease mortality: The Paris Prospective Study. *Arterioscler Thromb* 12: 1387–1392.
- Casolo G, Balli E, Taddei T, Amuhasi J & Gori C (1989) Decreased spontaneous heart rate variability in congestive heart failure. *Am J Cardiol* 64: 1162–1167.
- Chakko S, Mulingtapang RF, Huikuri HV, Kessler KM, Materson BJ & Myerburg RJ (1993) Alterations in heart rate variability and its circadian rhythm in hypertensive patients with left ventricular hypertrophy free of coronary artery disease. *Am Heart J* 126: 1364–1372.
- Cook JR, Bigger JT, Jr., Kleiger RE, Fleiss JL, Steinman RC & Rolnitzky LM (1991) Effect of atenolol and diltiazem on heart period variability in normal persons. *J Am Coll Cardiol* 17: 480–484.
- Cowan MJ, Pike K & Burr RL (1994) Effects of gender and age on heart rate variability in healthy individuals and in persons after sudden cardiac arrest. *J Electrocardiol* 27 Suppl: 1–9.
- Demirel S, Akkaya V, Oflaz H, Tukek T & Erk O (2002) Heart rate variability after coronary artery bypass graft surgery: a prospective 3-year follow-up study. *Ann Noninvasive Electrocardiol* 7: 247–250.
- Deprès JP & Marette A (1994) Relation of components of insulin resistance syndrome to coronary disease risk. *Curr Opin Lipidol* 5: 274–289.
- Dimmer C, Tavernier R, Gjorgov N, Van Nooten G, Clement DL & Jordaens L (1998) Variations of autonomic tone preceding onset of atrial fibrillation after coronary artery bypass grafting. *Am J Cardiol* 82: 22–25.
- Dzau VJ & Sacks FM (1987) Regulation of lipoprotein metabolism by adrenergic mechanisms. *J Cardiovasc Pharmacol* 10 Suppl 9: S2–S6.
- Eckberg DL (1983) Human sinus arrhythmia as an index of vagal cardiac outflow. *J Appl Physiol* 54: 961–966.
- Eckberg DL (1997) Sympathovagal balance: a critical appraisal. *Circulation* 96: 3224–3232.
- Eckberg DL, Drabinsky M & Braunwald E (1971) Defective cardiac parasympathetic control in patients with heart disease. *N Engl J Med* 285: 877–883.
- Ewing DJ, Neilson JMM & Travis P (1984) New method for assessing cardiac parasympathetic activity using 24 hours electrocardiograms. *Br Heart J* 52: 396–402.
- Facchini FS, Stoohs RA & Reaven GM (1996) Enhanced sympathetic nervous system activity. The linchpin between insulin resistance, hyperinsulinemia, and heart rate. *Am J Hypertens* 9: 1013–1017.
- Farrell TG, Odemuyiwa O, Bashir Y, Cripps TR, Malik M, Ward DE & Camm AJ (1992) Prognostic value of baroreflex sensitivity testing after acute myocardial infarction. *Br Heart J* 67: 129–137.

- Filipovsky J, Ducimetiere P & Safar ME (1992) Prognostic significance of exercise blood pressure and heart rate in middle-aged men. *Hypertension* 20: 333–339.
- Flapan AD, Wright RA, Nolan J, Neilson JM & Ewing DJ (1993) Differing patterns of cardiac parasympathetic activity and their evolution in selected patients with a first myocardial infarction. *J Am Coll Cardiol* 21: 926–931.
- Ford DE, Mead LA, Chang PP, Cooper-Patrick L, Wang NY & Klag M (1998) Depression is a risk factor for coronary artery disease in men: the precursor study. *Arch Intern Med*. 158: 1422–1426.
- Frick MH, Syväne M, Nieminen MS, Kauma H, Majahalme S, Virtanen V, Kesäniemi YA, Pasternack A & Taskinen MR (1997) Prevention of the angiographic progression of coronary and vein-graft atherosclerosis by gemfibrozil after coronary bypass surgery in men with low levels of HDL cholesterol. Lopid Coronary Angiography Trial (LOCAT) Study Group. *Circulation* 96: 2137–2143.
- Friedewald WT, Levy RI & Fredrickson DS (1972) Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem* 18: 499–502.
- Ghuran A, Reid F, La Rovere MT, Schmidt G, Bigger JT, Jr., Camm AJ, Schwartz PJ & Malik M (2002) Heart rate turbulence-based predictors of fatal and nonfatal cardiac arrest (The Autonomic Tone and Reflexes After Myocardial Infarction substudy). *Am J Cardiol* 89: 184–190.
- Gillum RF, Makuc DM & Feldman JJ (1991) Pulse rate, coronary heart disease, and death: the NHANES I Epidemiologic Follow-up Study. *Am Heart J* 121: 172–177.
- Goldberger AL (1996) Non-linear dynamics for clinicians: chaos theory, fractals, and complexity at the bedside. *Lancet* 347: 1312–1314.
- Goldberger AL & West BJ (1987) Applications of nonlinear dynamics to clinical cardiology. *Ann N Y Acad Sci* 504: 195–213.
- Grundy SM, Pasternak R, Greenland P, Smith S Jr & Fuster V (1999) Assessment of cardiovascular risk by use of multiple-risk-factor assessment equations: a statement for healthcare professionals from the American Heart Association and the American College of Cardiology. *Circulation* 100: 1481–1492.
- Haffner SM, Lehto S, Rönnemaa T, Pyörälä K & Laakso M (1998) Mortality from coronary heart disease in subjects with type 2 diabetes and in nondiabetic subjects with and without prior myocardial infarction. *N Engl J Med* 339: 229–234.
- Harris T, Cook EF, Kannel WB & Goldman L (1988) Proportional hazards analysis of risk factors for coronary heart disease in individuals aged 65 or older. The Framingham Heart Study. *J Am Geriatr Soc* 36: 1023–1028.
- Hartikainen JE, Malik M, Staunton A, Poloniecki J & Camm AJ (1996) Distinction between arrhythmic and nonarrhythmic death after acute myocardial infarction based on heart rate variability, signal-averaged electrocardiogram, ventricular arrhythmias and left ventricular ejection fraction. *J Am Coll Cardiol* 28: 296–304.
- Hayano J, Sakakibara Y, Yamada A, Yamada M, Mukai S, Fujinami T, Yokoyama K, Watanabe Y & Takata K (1991) Accuracy of assessment of cardiac vagal tone by heart rate variability in normal subjects. *Am J Cardiol* 67: 199–204.
- Hayano J, Sakakibara Y, Yamada M, Ohte N, Fujinami T, Yokoyama K, Watanabe Y & Takata K (1990) Decreased magnitude of heart rate spectral components in coronary artery disease. Its relation to angiographic severity. *Circulation* 81: 1217–1224.
- Ho KK, Moody GB, Peng CK, Mietus JE, Larson MG, Levy D & Goldberger AL (1997) Predicting survival in heart failure case and control subjects by use of fully automated methods for deriving nonlinear and conventional indices of heart rate dynamics. *Circulation* 96: 842–848.
- Hogue CW, Jr., Domitrovich PP, Stein PK, Despotis GD, Re L, Schuessler RB, Kleiger RE & Rottman JN (1998) RR interval dynamics before atrial fibrillation in patients after coronary artery bypass graft surgery. *Circulation* 98: 429–434.

- Hogue CW, Jr., Stein PK, Apostolidou I, Lappas DG & Kleiger RE (1994) Alterations in temporal patterns of heart rate variability after coronary artery bypass graft surgery. *Anesthesiology* 81: 1356–1364.
- Hohnloser SH, Klingenstein T, Zabel M, Schroder F & Just H (1992) Intraindividual reproducibility of heart rate variability. *Pacing Clin Electrophysiol* 15: 2211–2214.
- Huikuri HV, Kessler KM, Terracall E, Castellanos A, Linnaluoto MK & Myerburg RJ (1990) Reproducibility and circadian rhythm of heart rate variability in healthy subjects. *Am J Cardiol* 65: 391–393.
- Huikuri HV, Linnaluoto MK, Seppänen T, Airaksinen KE, Kessler KM, Takkunen JT & Myerburg RJ (1992) Circadian rhythm of heart rate variability in survivors of cardiac arrest. *Am J Cardiol* 70: 610–615.
- Huikuri HV, Mäkikallio TH, Airaksinen KE, Seppänen T, Puukka P, Räihä IJ & Sourander LB (1998) Power-law relationship of heart rate variability as a predictor of mortality in the elderly. *Circulation* 97: 2031–2036.
- Huikuri HV, Mäkikallio TH, Peng CK, Goldberger AL, Hintze U & Moller M (2000) Fractal correlation properties of R-R interval dynamics and mortality in patients with depressed left ventricular function after an acute myocardial infarction. *Circulation* 101: 47–53.
- Huikuri HV, Niemelä MJ, Ojala S, Rantala A, Ikäheimo MJ & Airaksinen KE (1994) Circadian rhythms of frequency domain measures of heart rate variability in healthy subjects and patients with coronary artery disease. Effects of arousal and upright posture. *Circulation* 90: 121–126.
- Huikuri HV, Ylitalo A, Pikkujämsä SM, Ikäheimo MJ, Airaksinen KE, Rantala AO, Lilja M & Kesäniemi YA (1996) Heart rate variability in systemic hypertension. *Am J Cardiol* 77: 1073–1077.
- Iellamo F, Legramante JM, Massaro M, Raimondi G & Galante A (2000) Effects of a residential exercise training on baroreflex sensitivity and heart rate variability in patients with coronary artery disease: A randomized, controlled study. *Circulation* 102: 2588–2592.
- Ismail A, Khosravi H & Olson H (1999) The role of infection in atherosclerosis and coronary artery disease: a new therapeutic target. *Heart Dis.* 1: 233–240.
- Iyengar N, Peng CK, Morin R, Goldberger AL & Lipsitz LA (1996) Age-related alterations in the fractal scaling of cardiac interbeat interval dynamics. *Am J Physiol* 271: R1078–R1084.
- Jost S, Deckers JW, Nikutta P, Wiese B, Rafflenbeul W, Hecker H, Lippolt P & Lichtlen PR (1994) Evolution of coronary stenoses is related to baseline severity-prospective quantitative angiographic analysis in patients with moderate coronary disease. *INTACT Investigators. International Nifedipine Trial on Antiatherosclerotic Therapy. Eur Heart J* 15: 648–653
- Jensen-Urstad K, Storck N, Bouvier F, Ericson M, Lindblad LE & Jensen-Urstad M (1997) Heart rate variability in healthy subjects is related to age and gender. *Acta Physiol Scand* 160: 235–241.
- Kannel WB, Kannel C, Paffenbarger RS, Jr. & Cupples LA (1987) Heart rate and cardiovascular mortality: the Framingham Study. *Am Heart J* 113: 1489–1494.
- Kaufman ES, Bosner MS, Bigger JT, Jr., Stein PK, Kleiger RE, Rolnitzky LM, Steinman RC & Fleiss JL (1993) Effects of digoxin and enalapril on heart period variability and response to head-up tilt in normal subjects. *Am J Cardiol* 72: 95–99.
- Kingwell BA, Cameron JD, Gillies KJ, Jennings GL & Dart AM (1995) Arterial compliance may influence baroreflex function in athletes and hypertensives. *Am J Physiol* 268: H411–H418
- Kleiger RE, Bigger JT, Bosner MS, Chung MK, Cook JR, Rolnitzky LM, Steinman R & Fleiss JL (1991) Stability over time of variables measuring heart rate variability in normal subjects. *Am J Cardiol* 68: 626–630.
- Kleiger RE, Miller JP, Bigger JT, Jr. & Moss AJ (1987) Decreased heart rate variability and its association with increased mortality after acute myocardial infarction. *Am J Cardiol* 59: 256–262.
- Kleiger RE, Stein PK, Bosner MS & Rottman JN (1992) Time domain measurements of heart rate variability. *Cardiol Clin* 10: 487–498.

- Korpelainen JT, Sotaniemi KA, Huikuri HV & Myllylä VV (1996) Abnormal heart rate variability as a manifestation of autonomic dysfunction in hemispheric brain infarction. *Stroke* 27: 2059–2063.
- Koskinen P, Mänttari M, Manninen V, Huttunen JK, Heinonen OP & Frick MH (1992) Coronary heart disease incidence in NIDDM patients in the Helsinki Heart Study. *Diabetes Care* 15: 820–825.
- Krumholz HM, Seeman TE, Merrill SS, Mendes de Leon CF, Vaccarino V, Silverman DI, Tsukahara R, Ostfeld AM & Berkman LF (1994) Lack of association between cholesterol and coronary heart disease mortality and morbidity and all-cause mortality in persons older than 70 years. *JAMA* 272: 1335–1340.
- Kukreja RS, Datta BN & Chakravarti RN (1981) Catecholamine-induced aggravation of aortic and coronary atherosclerosis in monkeys. *Atherosclerosis* 40: 291–298.
- Kupari M, Virolainen J, Koskinen P & Tikkanen MJ (1993) Short-term heart rate variability and factors modifying the risk of coronary artery disease in a population sample. *Am J Cardiol* 72: 897–903.
- Kuroiwa Y, Shimada Y & Toyokura Y (1983) Postural hypotension and low R-R interval variability in parkinsonism, spino-cerebellar degeneration, and Shy-Drager syndrome. *Neurology* 33: 463–467.
- La Rovere MT, Bigger JT, Jr., Marcus FI, Mortara A & Schwartz PJ (1998) Baroreflex sensitivity and heart-rate variability in prediction of total cardiac mortality after myocardial infarction. ATRAMI (Autonomic Tone and Reflexes After Myocardial Infarction) Investigators. *Lancet* 351: 478–484.
- La Rovere MT, Pinna GD, Maestri R, Mortara A, Capomolla S, Febo O, Ferrari R, Franchini M, Gnemmi M, Opasich C, Riccardi PG, Traversi E & Cobelli F (2003) Short-term heart rate variability strongly predicts sudden cardiac death in chronic heart failure patients. *Circulation* 107: 565–570.
- Laitinen T, Vauhkonen IK, Niskanen LK, Hartikainen JE, Länsimies EA, Uusitupa MI & Laakso M (1999) Power spectral analysis of heart rate variability during hyperinsulinemia in nondiabetic offspring of type 2 diabetic patients: evidence for possible early autonomic dysfunction in insulin-resistant subjects. *Diabetes* 48: 1295–1299.
- Laitio TT, Huikuri HV, Kentala ES, Mäkikallio TH, Jalonen JR, Helenius H, Sariola-Heinonen K, Yli-Mäyry S & Scheinin H (2000) Correlation properties and complexity of perioperative RR-interval dynamics in coronary artery bypass surgery patients. *Anesthesiology* 93: 69–80.
- Laitio TT, Mäkikallio TH, Huikuri HV, Kentala ES, Uotila P, Jalonen JR, Helenius H, Hartiala J, Yli-Mäyry S & Scheinin H (2002) Relation of heart rate dynamics to the occurrence of myocardial ischemia after coronary artery bypass grafting. *Am J Cardiol* 89: 1176–1181.
- Lesperance J & Waters D (1992) Measuring progression and regression of coronary atherosclerosis in clinical trials: problems and progress. *Int J Card Imaging* 8: 165–173.
- Lipsitz LA & Goldberger AL (1992) Loss of 'complexity' and aging. Potential applications of fractals and chaos theory to senescence. *JAMA* 267: 1806–1809.
- Lombardi F, Sandrone G, Pernpruner S, Sala R, Garimoldi M, Cerutti S, Baselli G, Pagani M & Malliani A (1987) Heart rate variability as an index of sympathovagal interaction after acute myocardial infarction. *Am J Cardiol* 60: 1239–1245.
- Lowensohn RI, Weiss M & Hon EH (1977) Heart-rate variability in brain-damaged adults. *Lancet* 1: 626–628.
- Mack WJ, Selzer RH, Pogoda JM, Lee PL, Shircore AM, Azen SP & Blankenhorn DH (1992) Comparison of computer- and human-derived coronary angiographic end-point measures for controlled trials. *Arterioscler Thromb* 12: 348–356.
- Madwed JB, Albrecht P, Mark RG & Cohen RJ (1989) Low-frequency oscillations in arterial pressure and heart rate: a simple computer model. *Am J Physiol* 256:H1573–H1579.

- Malpas SC, Whiteside EA & Maling TJ (1991) Heart rate variability and cardiac autonomic function in men with chronic alcohol dependence. *Br Heart J* 65: 84–88.
- Mattila K, Haavisto M, Rajala S & Heikinheimo R (1988) Blood pressure and five year survival in the very old. *Br Med J (Clin Res Ed)* 296: 887–889.
- McGuinness C, Seccombe DW, Frohlich JJ, Ehnholm C, Sundvall J & Steiner G (2000) Laboratory standardization of a large international clinical trial: the DAIS experience. DAIS Project Group. Diabetes Atherosclerosis Intervention Study. *Clin Biochem* 33: 15–24.
- McLaughlin PR & Gladstone P (1998) Diabetes atherosclerosis intervention study (DAIS): quantitative coronary angiographic analysis of coronary artery atherosclerosis. *Cathet Cardiovasc Diagn* 44: 249–256.
- Mensink GB & Hoffmeister H (1997) The relationship between resting heart rate and all-cause, cardiovascular and cancer mortality. *Eur Heart J* 18: 1404–1410.
- Molgaard H, Mickley H, Pless P, Bjerregaard P & Moller M (1993) Effects of metoprolol on heart rate variability in survivors of acute myocardial infarction. *Am J Cardiol* 71: 1357–1359.
- Montano N, Ruscone TG, Porta A, Lombardi F, Pagani M & Malliani A (1994) Power spectrum analysis of heart rate variability to assess the changes in sympathovagal balance during graded orthostatic tilt. *Circulation* 90: 1826–1831.
- Mortara A, La Rovere MT, Pinna GD, Maestri R, Capomolla S & Cobelli F (2000) Nonselective beta-adrenergic blocking agent, carvedilol, improves arterial baroflex gain and heart rate variability in patients with stable chronic heart failure. *J Am Coll Cardiol* 36: 1612–1618.
- Multiple Risk Factor Intervention Trial (1996) Mortality after 16 years for participants randomized to the Multiple Risk Factor Intervention Trial. *Circulation* 94: 946–951.
- Mäkikallio TH, Hoiber S, Kober L, Torp-Pedersen C, Peng CK, Goldberger AL & Huikuri HV (1999a) Fractal analysis of heart rate dynamics as a predictor of mortality in patients with depressed left ventricular function after acute myocardial infarction. TRACE Investigators. TRAndolapril Cardiac Evaluation. *Am J Cardiol* 83: 836–839.
- Mäkikallio TH, Huikuri HV, Hintze U, Videbaek J, Mitrani RD, Castellanos A, Myerburg RJ & Moller M (2001a) Fractal analysis and time- and frequency-domain measures of heart rate variability as predictors of mortality in patients with heart failure. *Am J Cardiol* 87: 178–182.
- Mäkikallio TH, Huikuri HV, Mäkikallio A, Sourander LB, Mitrani RD, Castellanos A & Myerburg RJ (2001b) Prediction of sudden cardiac death by fractal analysis of heart rate variability in elderly subjects. *J Am Coll Cardiol* 37: 1395–1402.
- Mäkikallio TH, Koistinen J, Jordaens L, Tulppo MP, Wood N, Golosarsky B, Peng CK, Goldberger AL & Huikuri HV (1999b) Heart rate dynamics before spontaneous onset of ventricular fibrillation in patients with healed myocardial infarcts. *Am J Cardiol* 83: 880–884.
- Mäkikallio TH, Ristimäe T, Airaksinen KE, Peng CK, Goldberger AL & Huikuri HV (1998) Heart rate dynamics in patients with stable angina pectoris and utility of fractal and complexity measures. *Am J Cardiol* 81: 27–31.
- Mäkikallio TH, Seppänen T, Niemelä M, Airaksinen KE, Tulppo M & Huikuri HV (1996) Abnormalities in beat to beat complexity of heart rate dynamics in patients with a previous myocardial infarction. *J Am Coll Cardiol* 28: 1005–1011.
- Niemelä MJ, Airaksinen KE & Huikuri HV (1994) Effect of beta-blockade on heart rate variability in patients with coronary artery disease. *J Am Coll Cardiol* 23: 1370–1377.
- Niemelä MJ, Airaksinen KE, Tahvanainen KU, Linnaluoto MK & Takkunen JT (1992) Effect of coronary artery bypass grafting on cardiac parasympathetic nervous function. *Eur Heart J* 13: 932–935.
- Odemuyiwa O, Malik M, Farrell T, Bashir Y, Poloniecki J & Camm J (1991) Comparison of the predictive characteristics of heart rate variability index and left ventricular ejection fraction for all-cause mortality, arrhythmic events and sudden death after acute myocardial infarction. *Am J Cardiol* 68: 434–439.

- Pagani M, Lombardi F, Guzzetti S, Rimoldi O, Furlan R, Pizzinelli P, Sandrone G, Malfatto G, Dell'Orto S, Piccaluga E & . (1986) Power spectral analysis of heart rate and arterial pressure variabilities as a marker of sympatho-vagal interaction in man and conscious dog. *Circ Res* 59: 178–193.
- Peng CK, Havlin S, Stanley HE & Goldberger AL (1995) Quantification of scaling exponents and crossover phenomena in nonstationary heartbeat time series. *Chaos* 5: 82–87.
- Peng CK, Mietus JE, Liu Y, Lee C, Hausdorff JM, Stanley HE, Goldberger AL & Lipsitz LA (2002) Quantifying fractal dynamics of human respiration: age and gender effects. *Ann Biomed Eng* 30: 683–692.
- Perkiömäki JS, Huikuri HV, Koistinen JM, Mäkikallio T, Castellanos A & Myerburg RJ (1997) Heart rate variability and dispersion of QT interval in patients with vulnerability to ventricular tachycardia and ventricular fibrillation after previous myocardial infarction. *J Am Coll Cardiol* 30: 1331–1338.
- Perkiömäki JS, Zareba W, Kalaria VG, Couderc J-P, Huikuri HV & Moss AJ (2001a) Comparability of nonlinear measures of heart rate variability between long- and short-term electrocardiographic recordings. *Am J Cardiol* 87: 905–908.
- Perkiömäki JS, Zareba W, Ruta J, Dubner S, Madoery C, Deedwania P, Karcz M & Bayes de Luna A (2001b) Fractal and complexity measures of heart rate dynamics after acute myocardial infarction. *Am J Cardiol* 88: 777–781.
- Perski A, Olsson G, Landou C, de Faire U, Theorell T & Hamsten A (1992) Minimum heart rate and coronary atherosclerosis: independent relations to global severity and rate of progression of angiographic lesions in men with myocardial infarction at a young age. *Am Heart J* 123: 609–616.
- Pikkujämsä SM, Huikuri HV, Airaksinen KE, Rantala AO, Kauma H, Lilja M, Savolainen MJ & Kesäniemi YA (1998) Heart rate variability and baroreflex sensitivity in hypertensive subjects with and without metabolic features of insulin resistance syndrome. *Am J Hypertens* 11: 523–531.
- Pikkujämsä SM, Mäkikallio TH, Airaksinen KE & Huikuri HV (2001) Determinants and interindividual variation of R-R interval dynamics in healthy middle-aged subjects. *Am J Physiol Heart Circ Physiol* 280: H1400–H1406.
- Pikkujämsä SM, Mäkikallio TH, Sourander LB, Räihä IJ, Puukka P, Skyttä J, Peng CK, Goldberger AL & Huikuri HV (1999) Cardiac interbeat interval dynamics from childhood to senescence: comparison of conventional and new measures based on fractals and chaos theory. *Circulation* 100: 393–399.
- Pincus SM (1991) Approximate entropy as a measure of system complexity. *Proc Natl Acad Sci U S A* 88: 2297–2301.
- Pincus SM & Goldberger AL (1994) Physiological time-series analysis: what does regularity quantify? *Am J Physiol* 266: H1643–H1656.
- Pincus SM & Viscarello RR (1992) Approximate entropy: a regularity measure for fetal heart rate analysis. *Obstet Gynecol* 79: 249–255.
- Pomeranz B, Macaulay RJ, Caudill MA, Kutz I, Adam D, Gordon D, Kilborn KM, Barger AC, Shannon DC, Cohen RJ & . (1985) Assessment of autonomic function in humans by heart rate spectral analysis. *Am J Physiol* 248: H151–H153.
- Pyörälä K, Savolainen E, Kaukola S & Haapakoski L (1985) Plasma insulin as coronary heart disease risk factor: relationship to other risk factors and predictive value during 9.5-year follow-up of the Helsinki Policeman Study population. *Acta Med Scand* 701(suppl 1): 38–52.
- Quinn TG, Alderman EL, Mcmillan A & Haskell W (1994) Development of new coronary atherosclerotic lesions during a 4-year multifactor risk reduction program: the Stanford Coronary Risk Intervention project (SCRIP). *J Am Coll Cardiol* 24: 900–908.
- Rathmann W, Ziegler D, Jahnke M, Haastert B & Gries FA (1993) Mortality in diabetic patients with cardiovascular autonomic neuropathy. *Diabet Med* 10: 820–824.

- Reaven GM, Lithell H & Landsberg L (1996) Hypertension and associated metabolic abnormalities—the role of insulin resistance and the sympathoadrenal system. *N Engl J Med* 334: 374–381.
- Rubins HB, Robins SJ, Collins D, Fye CL, Anderson JW, Elam MB, Faas FH, Linares E, Schaefer EJ, Schectman G, Wilt TJ & Wittes J (1999) Gemfibrozil for the secondary prevention of coronary heart disease in men with low levels of high-density lipoprotein cholesterol. Veterans Affairs High-Density Lipoprotein Cholesterol Intervention Trial Study Group. *N Engl J Med* 341: 410–418.
- Räihä JJ, Marniemi J, Puukka P, Toikka T, Enholm C & Sourander LB (1997) Effect of serum lipids, lipoproteins, and apolipoproteins on vascular and non-vascular mortality in the elderly. *Arterioscler Thromb Vasc Biol.* 17: 1224–1232
- Sandrone G, Mortara A, Torzillo D, La Rovere MT, Malliani A & Lombardi F (1994) Effects of beta blockers (atenolol or metoprolol) on heart rate variability after acute myocardial infarction. *Am J Cardiol* 74: 340–345.
- Sands KE, Appel ML, Lilly LS, Schoen FJ, Mudge GH, Jr. & Cohen RJ (1989) Power spectrum analysis of heart rate variability in human cardiac transplant recipients. *Circulation* 79: 76–82.
- Sayers BM (1973) Analysis of heart rate variability. *Ergonomics* 16: 17–32.
- Schmidt G, Malik M, Barthel P, Schneider R, Ulm K, Rolnitzky L, Camm AJ, Bigger JT, Jr. & Schomig A (1999) Heart-rate turbulence after ventricular premature beats as a predictor of mortality after acute myocardial infarction. *Lancet* 353: 1390–1396.
- Shannon DC, Carley DW & Benson H (1987) Aging of modulation of heart rate. *Am J Physiol* 253: H874–H877.
- Solberg LA & Strong JP (1983) Risk factors and atherosclerotic lesions. A review of autopsy studies. *Arteriosclerosis* 3: 187–198.
- Stamler J, Daviglius ML, Garside DB, Dyer AR, Greenland P & Neaton JD (2000) Relationship of baseline serum cholesterol levels in 3 large cohorts of younger men to long-term coronary, cardiovascular, and all-cause mortality and to longevity. *JAMA* 284: 311–318.
- Stamler J, Vaccaro O, Neaton JD & Wentworth D (1993) Diabetes, other risk factors, and 12-yr cardiovascular mortality for men screened in the Multiple Risk Factor Intervention Trial. *Diabetes Care* 16: 434–444.
- Stein PK, Domitrovich PP, Kleiger RE, Schechtman KB & Rottman JN (2000) Clinical and demographic determinants of heart rate variability in patients post myocardial infarction: insights from the cardiac arrhythmia suppression trial (CAST). *Clin Cardiol* 23: 187–194.
- Stone PH, Gibson CM, Pasternak RC, Mcmanus K, Diaz L, Boucher T, Spears R, Sandor T, Rosner B & Sacks FM (1993) Natural history of coronary atherosclerosis using quantitative angiography in men, and implications for clinical trials of coronary regression. The Harvard Atherosclerosis Reversibility Project Study Group. *Am J Cardiol* 71: 766–772.
- Stern MP, Morales PA, Haffner SM & Valdez RA (1992) Hyperdynamic circulation and the insulin resistance syndrome ("syndrome X"). *Hypertension* 20: 802–808.
- Suda Y, Otsuka K, Niinami H, Ichikawa S, Ban T, Higashita R & Takeuchi Y (2001) Changes in ultra-low and very low frequency heart rate variability after coronary artery bypass grafting. *Biomed Pharmacother* 55 Suppl 1: 110s–114s.
- Syvänne M, Taskinen M-R, Nieminen MS, Manninen V, Kesäniemi YA, Pasternack A, Nawrocki JW, Haber H & Frick MH (1997) A study to determine the response of coronary atherosclerosis to raising low HDL cholesterol with a fibric-acid derivative in men after coronary bypass surgery: the rationale, design and baseline characteristics of the locat study. *Control Clin. Trials.* 18: 93–119.
- Syvänne M, Nieminen MS & Frick MH (1994) Accuracy and precision of quantitative arteriography in the evaluation of coronary artery disease after coronary bypass surgery. *Int J Card Imaging* 10: 241–242

- Tapanainen JM, Thomsen PE, Kober L, Torp-Pedersen C, Mäkikallio TH, Still AM, Lindgren KS & Huikuri HV (2002) Fractal analysis of heart rate variability and mortality after an acute myocardial infarction. *Am J Cardiol* 90: 347–352.
- Tasaki H, Serita T, Irita A, Hano O, Iliev I, Ueyama C, Kitano K, Seto S, Hayano M & Yano K (2000) A 15-year longitudinal follow-up study of heart rate and heart rate variability in healthy elderly persons. *J Gerontol A Biol Sci Med Sci* 55: M744–M749.
- Task Force (1996) 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* 93: 1043–1065.
- Taylor JA, Carr DL, Myers CW & Eckberg DL (1998) Mechanisms underlying very-low-frequency RR-interval oscillations in humans. *Circulation* 98: 547–555.
- Töyry JP, Niskanen LK, Mäntysaari MJ, Länsimies EA, Haffner SM, Miettinen HJ & Uusitupa MI (1997) Do high proinsulin and C-peptide levels play a role in autonomic nervous dysfunction?: Power spectral analysis in patients with non-insulin-dependent diabetes and nondiabetic subjects. *Circulation* 96: 1185–1191.
- Töyry JP, Niskanen LK, Mäntysaari MJ, Länsimies EA & Uusitupa MI (1996) Occurrence, predictors, and clinical significance of autonomic neuropathy in NIDDM. Ten-year follow-up from the diagnosis. *Diabetes* 45: 308–315.
- Troisi RJ, Weiss ST, Parker DR, Sparrow D, Young JB & Landsberg L (1991) Relation of obesity and diet to sympathetic nervous system activity. *Hypertension* 17: 669–677.
- Tsuji H, Larson MG, Venditti FJ, Jr., Manders ES, Evans JC, Feldman CL & Levy D (1996) Impact of reduced heart rate variability on risk for cardiac events. The Framingham Heart Study. *Circulation* 94: 2850–2855.
- Tsuji H, Venditti FJ, Jr., Manders ES, Evans JC, Larson MG, Feldman CL & Levy D (1994) Reduced heart rate variability and mortality risk in an elderly cohort. The Framingham Heart Study. *Circulation* 90: 878–883.
- Tulppo MP, Mäkikallio TH, Seppänen T, Shoemaker K, Tutungi E, Hughson RL & Huikuri HV (2001) Effects of pharmacological adrenergic and vagal modulation on fractal heart rate dynamics. *Clin Physiol* 21: 515–523.
- Umetani K, Singer DH, McCraty R & Atkinson M (1998) Twenty-four hour time domain heart rate variability and heart rate: relations to age and gender over nine decades. *J Am Coll Cardiol* 31: 593–601.
- Vaage-Nilsen M, Rasmussen V, Jensen G, Simonsen L & Mortensen LS (2001) Recovery of autonomic nervous activity after myocardial infarction demonstrated by short-term measurements of SDNN. *Scand Cardiovasc J* 35: 186–191.
- van de Borne P, Montano N, Pagani M, Oren R & Somers VK (1997) Absence of low-frequency variability of sympathetic nerve activity in severe heart failure. *Circulation* 95: 1449–1454.
- Van Hoogenhuyze D, Weinstein N, Martin GJ, Weiss JS, Schaad JW, Sahyouni XN, Fintel D, Remme WJ & Singer DH (1991) Reproducibility and relation to mean heart rate of heart rate variability in normal subjects and in patients with congestive heart failure secondary to coronary artery disease. *Am J Cardiol* 68: 1668–1676.
- Vartiainen E, Jousilahti E, Alfthan G, Sundvall J, Pietinen P & Puska P (2000) Cardiovascular risk factor changes in Finland, 1972–1997. *Int J Epidemiol* 29: 49–56.
- Vartiainen E, Laatikainen T, Tapanainen H, Salomaa V, Jousilahti P, Männistö S, Valsta L, Sundvall J & Salminen I (2003) Changes in cardiovascular risk factors in Finland in the National FINNRISK Study between 1982 and 2002. *Finnish Medical Journal* 41: 4099–4106.
- Vikman S, Mäkikallio TH, Yli-Mäyry S, Pikkujämsä S, Koivisto AM, Reinikainen P, Airaksinen KE & Huikuri HV (1999) Altered complexity and correlation properties of R-R interval dynamics before the spontaneous onset of paroxysmal atrial fibrillation. *Circulation* 100: 2079–2084.

- Vita G, Bellinghieri G, Trusso A, Costantino G, Santoro D, Monteleone F, Messina C & Savica V (1999) Uremic autonomic neuropathy studied by spectral analysis of heart rate. *Kidney Int* 56: 232–237.
- Waters D, Lesperance J, Craven TE, Hudon G & Gillam LD (1993) Advantages and limitations of serial coronary arteriography for the assessment of progression and regression of coronary atherosclerosis. Implications for clinical trials. *Circulation* 87(3 Suppl): II38–47).
- Wexler L, Brundage B, Crouse J, Detrano R, Fuster V, Maddahi J, Rumberger J, Stanford W, White R & Taubert K (1996) Coronary artery calcification: pathophysiology, epidemiology, imaging methods, and clinical implications. A statement for health professionals from the American Heart Association. Writing Group. *Circulation* 94: 1175–1192.
- Yu SM, Tsai SY, Guh JH, Ko FN, Teng CM & Ou JT (1996) Mechanism of catecholamine-induced proliferation of vascular smooth muscle cells. *Circulation* 94: 547–554.
- Yusuf S, Peto R, Lewis J, Collins R & Sleight P (1985) Beta blockade during and after myocardial infarction: an overview of the randomized trials. *Prog Cardiovasc Dis* 27: 335–371.
- Zuanetti G, Latini R, Maggioni AP, Santoro L & Franzosi MG (1993) Influence of diabetes on mortality in acute myocardial infarction: data from the GISSI-2 study. *J Am Coll Cardiol* 22: 1788–1794.
- Zuanetti G, Latini R, Neilson JM, Schwartz PJ & Ewing DJ (1991) Heart rate variability in patients with ventricular arrhythmias: effect of antiarrhythmic drugs. Antiarrhythmic Drug Evaluation Group (ADEG). *J Am Coll Cardiol* 17: 604–612.