Age and gender differences in risk factor burden, myocardial dysfunction and cardiovascular events in relation to glucose metabolism

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“Not everything that counts can be counted,
and not everything that can be counted counts.”
William Bruce Cameron

CONTENTS

ABSTRACT

The risk of cardiovascular disease (CVD) and heart failure (HF) among individuals with diabetes is at least two times greater than in non-diabetic subjects. However, the excess risk of CVD in diabetic subjects seems to decrease with age. As the majority of patients with diabetes, CVD and HF are elderly it is important to establish the extent to which the associations between these conditions differ from those in younger populations. The overall aim of the work presented in this thesis was to study gender-related associations between glucose metabolism and myocardial dysfunction, risk factor burden and CVD events in middle-aged and elderly subjects. The hypothesis tested was that similar associations would be observed in elderly as in younger populations, although the associations would be weaker with advancing age. Data from two population-based cohort studies were used: MPP-RES (Sweden: n=18,238, mean age 69±6 years, range 57-86 years) and AGES-RS (Iceland: n=5,764, mean age 76±6 years, range 67-95).

In Paper I the associations between echocardiographic indices of left ventricular diastolic dysfunction (LVDD), LV mass index (LVMI) and glucometabolic status were studied in echocardiography subcohorts from the two cohort studies (MPP-RES n=1,792; AGES-RS n=841). The MPP-RES cohort was divided into two age groups: middle-aged (57-69 years) and elderly subjects (70-80 years). All subjects were grouped according to fasting glucose level (FG, mmol/l): ≤5.0; 5.1-5.5; 5.6-6.0 and 6.1-6.9 (pre-diabetic range) and ≥7.0 (new-onset diabetes) and established diabetes, and trends between the groups were assessed. Few and inconsistent associations were observed in the AGES-RS cohort and in the elderly group in the MPP-RES cohort between increasing glucometabolic impairment and measures of LVDD. These observations are in contrast to previous findings in younger subjects as well as the present findings in the middle-aged group in the MPP-RES cohort, where a significant association was found between increasing LVDD and increasing glucometabolic impairment. It was concluded that changes in LV diastolic function may be more related to age than glucose metabolism in elderly subjects.

In Paper II possible associations between N-terminal pro-B-type natriuretic peptide (Nt-proBNP) and FG as a continuous variable and in FG groups (as in Paper I) were assessed in the MPP-RES echocardiography subcohort. A positive correlation was found between Nt-proBNP and FG among middle-aged men. A positive correlation was also observed among elderly men, albeit non-significant. A non-significant negative correlation was observed among women in both age groups. The results indicate that caution should probably be exercised when interpreting Nt-proBNP values in subjects with impaired glucose metabolism.

In Paper III the strength of the correlation between glucometabolic impairment, CVD risk factor burden and self-rated health (SRH), in middle-aged and elderly groups in the whole MPP-RES cohort (n=18,238) was compared. Correlations between increasing glucometabolic impairment and CVD risk factor burden and the proportion of subjects reporting poor SRH increased for both men and women in both age groups (p-trend <0.0001 for all). The slope of the trend curve with increasing CVD risk factor burden was significantly steeper for elderly women than for elderly men (p-interaction=0.002). The slope of the trend curve for poor SRH was significantly steeper for middle-aged than for elderly men (p-interaction=0.005), while no difference was observed between the age groups in the women. These results indicate lifelong CVD risk factor clustering and poorer SRH with increased glucometabolic impairment, being somewhat more pronounced in elderly women than in elderly men.

Finally, in Paper IV we examined whether the previously observed age-related reduction in excess CVD risk for diabetic compared to non-diabetic subjects also applies to pre-diabetic conditions. The MPP-RES cohort was followed for 4.1±1.3 years during which 1,296 CVD events occurred. Subjects were grouped by FG, gender and age, as previously. The hazard ratios for CVD events increased with increasing FG among middle-aged men and women. No comparable increase was observed among elderly subjects, where men with FG ≤5.0-6.9 and women with FG ≤5.0-6.0 had HRs close to 1.0. The β-coefficients for interaction between age groups and intergroup trends were -0.17 (unadjusted p=0.01) and -0.15 (fully adjusted p=0.03) for men, and -0.13 (p=0.27) for women (irrespective of adjustment). It was concluded that FG values within the upper pre-diabetic range conveyed less excess risk of CVD events among elderly than middle-aged men and women.

ABBREVIATIONS

AGES-RS	Age Gene/Environment Susceptibility Reykjavik Study
BMI	Body mass index
BNP	B-type natriuretic peptide
CHD	Coronary heart disease
CVD	Cardiovascular disease
DBP	Diastolic blood pressure
ECG	Electrocardiogram
FG	Fasting glucose
FPG	Fasting plasma glucose
HDL	High-density lipoprotein
HF	Heart failure
HR	Hazard ratio
IFG	Impaired fasting glucose
IGT	Impaired glucose tolerance
LA	Left atrium
LDL	Low-density lipoprotein
LVDD	Left ventricular diastolic dysfunction
LVEF	Left ventricular ejection fraction
LVH	Left ventricular hypertrophy
LVMI	Left ventricular mass index
LVSD	Left ventricular systolic dysfunction

MI	Myocardial infarction
MPP-RES	Malmö Preventive Project Re-Examination Study
NFG	Normal fasting glucose
NP	Natriuretic peptide
Nt-proBNP	N-terminal pro B-type natriuretic peptide
OGTT	Oral glucose tolerance test
OR	Odds ratio
SBP	Systolic blood pressure
SD	Standard deviation
SRH	Self-rated health
TDI	Tissue Doppler imaging
TG	Triglycerides

LIST OF PAPERS

This thesis is based on the following papers, referred to hereafter by their Roman numerals:

I Leosdottir M, Willenheimer R, Plehn J, Borgquist R, Gudmundsson P, Harris TB, Launer L J, Bjornsdottir H, Nilsson P M, Gudnason V
Myocardial structure and function by echocardiography in relation to glucometabolic status in elderly subjects from 2 population-based cohorts: a cross-sectional study
Am Heart J. 2010;159(3):414-420.e4

II Leosdottir M, Willenheimer R, Hall C, Tjora S, Malm J, Melander O, Nilsson P M
Age and gender differences in the association between Nt-proBNP and glucometabolic disturbances
Scand Cardiovasc J. 2011;45(5):294-300

III Leosdottir M, Willenheimer R, Persson M, Nilsson P M
The association between glucometabolic disturbances, traditional cardiovascular risk factors and self-rated health by age and gender: a cross-sectional analysis within the Malmö Preventive Project
Cardiovasc Diabetol. 2011;10(1):118

IV Leosdottir M, Willenheimer R, Nilsson P M
The influcence of age and gender on cardiovascular event risk in relation to hyperglycaemia - The Malmö Preventive Project Re-examination Study (MPP-RES)
Manuscript

BACKGROUND

Although the incidence of cardiovascular disease (CVD) has decreased in Western societies during recent decades, it is still the leading cause of mortality worldwide, accounting for 30% of deaths globally and 50% in Europe.[1] Mortality rates for heart failure (HF) have declined during the same period, also in Sweden.[2, 3] Life expectancy continues to increase in all parts of the world and, consequently, the elderly population is expanding (Figure 1).[4] As the incidence of both CVD and HF increases with age, an increase in the prevalence of CVD and HF, and a corresponding increase in medical care costs, can be expected.[5]

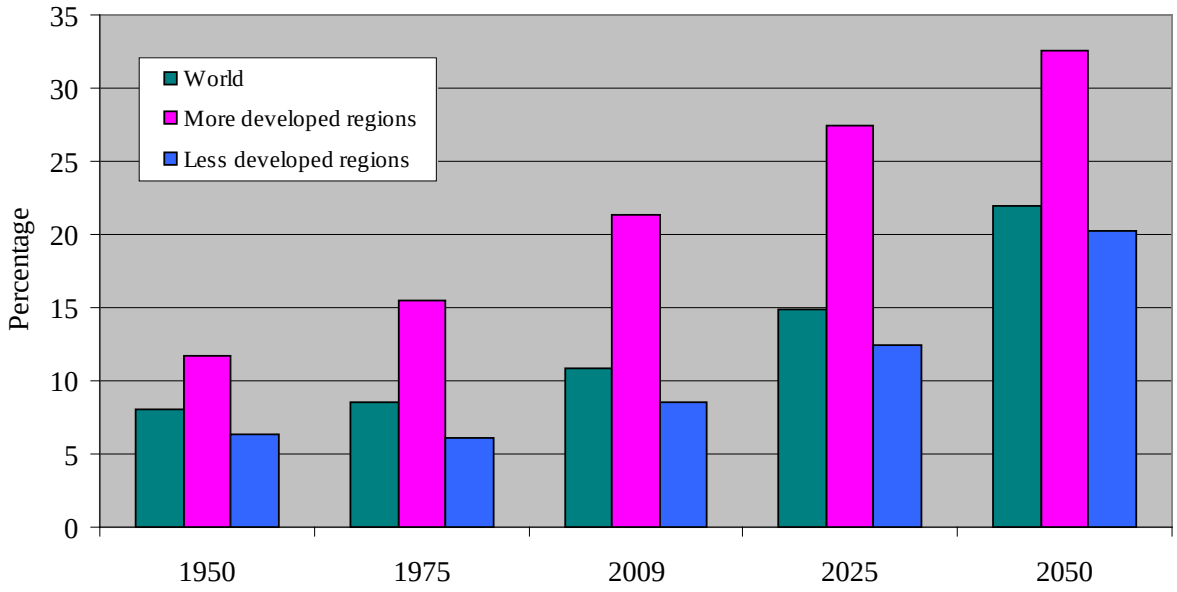

Figure 1. United Nations' projections for the percentage of the world population aged 60 or over.[4] © United Nations 2009.

The INTERHEART[6] and INTERSTROKE[7] studies showed that nine or ten modifiable risk factors account for as much as 90% of the population-attributable risk of myocardial infarction (MI) and stroke. Model estimates have suggested that approximately half of the decline in CVD mortality observed in industrialized countries during the past thirty years is due to primary and secondary prevention.[8, 9] Screening for, and treating risk factors for CVD, especially among high-risk populations, should therefore be given high priority.[10] Subjects with impaired glucose metabolism belong to this group. The risk of CVD and HF among individuals with diabetes is 2-5 times higher than in non-diabetic subjects; women bearing a disproportionate burden of the risk.[11]

Cardiovascular disease

Cardiovascular disease constitutes a group of disorders in which atherosclerosis is the common pathological denominator. Atherosclerosis leads to ischaemia, which may cause damage to the organs supplied by the affected arteries. CVD is by far the most common cause of death in Europe. According to statistics presented by the World Health Organization (WHO) CVD caused 50% of deaths in Europe in 2008, followed by cancer, which caused 20% of all deaths.[1] Although the incidence is falling, especially in Northern and Western Europe, the prevalence of CVD and the societal disease burden remain high, with 34 million disability-adjusted life years lost each year.[12] Coronary heart disease (CHD) is the largest contributor to CVD mortality throughout the world, followed by cerebrovascular disease, causing ischaemic stroke.[1, 13]

CVD affects different organ systems in the body, but principally the same patho-physiology and risk factors are at work.[14] Apart from non-modifiable risk factors such as age, gender and family history, the most important risk factors are hypertension, smoking, dyslipidaemia, diabetes, unfavourable psychosocial and socio-economic status and unhealthy lifestyle including physical inactivity, poor dietary habits and obesity.[6, 7, 14]

Heart failure

Heart failure is a pathophysiological state in which the capacity of the heart cannot meet the body's needs. HF has many underlying causes, of which CHD and hypertension are the most common; the less common ones being cardiomyopathy, valvular disease, cardiac arrhythmias, alcohol overconsumption and diabetes.[15, 16] The prevalence of HF increases sharply with age, being approximately 2-3% in the general population and 10-20% among those aged 70 years and older.[16] Comorbidity and hospitalization are common and the prognosis is poor, with a 5-year mortality rate of approximately 50%.[17] Health care costs for the treatment of HF are high, representing 1-2% of the total health care budget in most developed countries.[18]

The functional changes in the heart that accompany symptomatic HF are systolic and/or diastolic myocardial dysfunction. Diastolic dysfunction includes the inability of the heart muscle to properly fill during diastole (the filling phase), while systolic dysfunction is characterized by inefficient pumping capacity during systole (the emptying phase). These are often seen in conjunction with, or are preceded by, structural changes including left ventricular hypertrophy (LVH) (especially in the presence of hypertension) and dilation of one or more of the cardiac chambers. These structural changes are sometimes referred to as cardiac or ventricular remodelling (when primarily affecting the LV).[19] All of these structural and functional changes can be present for some years before symptoms appear, and are associated with a poor

prognosis, indicating the need for awareness of the risk, prompt diagnosis and treatment in vulnerable populations.[20]

Impaired glucose metabolism

Epidemiology

Diabetes mellitus is a metabolic disorder characterized by hyperglycaemia (high blood sugar), which in turn is caused by inadequate insulin production, increased insulin resistance or both. Excluding the rarer forms, diabetes is divided into two main subtypes, type 1 and type 2. Type 1 typically develops in early life and is characterized by the autoimmune destruction of the insulin-producing beta cells of the pancreas, leading to total dependency on exogenous insulin. Type 2 usually appears later in life and is characterized by insulin resistance and relative insulin deficiency. Additionally, type 2 diabetes is often seen together with abdominal obesity, hypertension and lipid derangements, or dyslipidaemia (low high-density lipoprotein (HDL) and high triglycerides (TG)), jointly referred to as the metabolic syndrome.[21] Type 2 diabetes accounts for approximately 90% of diabetes cases worldwide.[22] The lifetime risk of diabetes is approximately the same in men and women and, as with CVD and HF, the incidence increases with age. The prevalence varies greatly between populations. In the USA the prevalence of type 2 diabetes has escalated to epidemic proportions[23], while recent studies in Sweden indicate a slowly increasing prevalence, with an incidence rate that is stagnating or even declining.[24, 25] On the global scale, however, prevalence of type 2 diabetes is increasing rapidly due to the considerable increase in the developing countries. The worldwide prevalence of diabetes in those over 20 years of age was estimated to be 4.6% in 2000 and projected to rise to 6.4% by the year 2030.[26] At least half as many again are believed to have undiagnosed diabetes, and twice as many have impaired fasting glucose (IFG) or impaired glucose tolerance (IGT), which will lead to diabetes in more than 60% of cases if no measures are taken.[22] Suggested explanations for the increase in prevalence include ageing populations, a general increase in body weight, a more sedentary lifestyle, better diagnostics, lower diagnostic thresholds and earlier onset of the disease.[27, 28]

Definitions and diagnosis

In 2006 the WHO and the International Diabetes Federation issued a revised consensus statement on the definition and diagnosis of diabetes (type 1 and type 2).[29] A fasting plasma glucose level (FPG) of≥7.0 mmol/l or plasma glucose of ≥11.1 mmol/l two hours after an oral glucose challenge (referred to as an oral glucose tolerance test, OGTT) are recommended as diagnostic cut-off values. These values are based on the correlation between FPG and the incidence of diabetic retinopathy, which increases dramatically around 7.0 mmol/l.[30, 31] The incidence of CVD (often referred to as macrovascular complications) however, seems to increase in a linear or J-shaped fashion with increasing FPG without any apparent threshold.[32, 33] According to the WHO definition FPG <6.1 mmol/l is regarded as normal, and 6.1-6.9 mmol/l is

defined as IFG. IGT is defined as a 2-hour plasma glucose level of 7.8-11.1 mmol/l. Women are more likely to have isolated IGT, while IFG is more common in men.[34, 35] An OGTT is recommended by the WHO in subjects with IFG, as FPG fails to diagnose at least 30% of diabetic subjects.[36] This proportion is even higher in elderly individuals, as described by Barrett-Connor et al., who found that 70% of women and 48% of men aged 50-89 years were diagnosed with diabetes according to 2-hour plasma glucose values but not by FPG.[37] The American Diabetes Association (ADA) lowered the threshold for IFG to 5.6 mmol/l in 2003,[38] but the WHO concluded in 2006 that there was insufficient scientific evidence for such a revision.[29]

The measurement of glycosylated haemoglobin A1c (HbA1c), first introduced in the 1970s, is the most widely used method for monitoring glycaemic control in diabetic subjects.[39, 40] HbA1c is formed by an irreversible interaction between glucose and amino group residues in haemoglobin and reflects the average level of blood glucose over the previous 2-3 months.[41] HbA1c correlates well with micro- and macrovascular complications,[42, 43] and has been suggested not only as a means of monitoring, but also diagnosing, diabetes.[44] Indeed, the ADA included the measurement of HbA1c as a means of diagnosing diabetes in 2010, recommending a cut-off value of ≥6.5% (DCCT/NGSP standard).[45, 46] The WHO has been hesitant, arguing that different measurement methods, availability and cost limit its use for diagnostic purposes.[29] Recently, a global measurement standard has been set by the International Federation of Clinical Chemistry making HbA1c measurements comparable worldwide.[47] This should facilitate the use of HbA1c as a diagnostic tool for diabetes in the near future.

IFG and diabetes, and IGT where applicable, will hereafter be jointly referred to as *impaired glucose metabolism.*

The association between CVD, HF and impaired glucose metabolism

The association between glycosuria and CHD was first described by the American cardiologist Samuel A. Levine in 1922.[48] Since then, diabetes has been well established as a risk factor for CVD and HF.[11, 49, 50] Over 70% of the total mortality among diabetic subjects is attributable to CVD, compared to 50% in the general population.[1, 51, 52] Not only established diabetes, but also fasting glucose (FG) below the diabetes threshold and IGT have been shown to be risk factors for CVD.and HF [32, 33, 53] The explanation for this lies partly in the fact that subjects with impaired glucose metabolism have more concomitant CVD risk factors, where hypertension with secondary development of LVH probably contributes most,[54] with LVH being a strong predictor of CVD incidence and mortality.[55-57] Macro- and microvascular atherosclerosis are also more common in subjects with diabetes.[58, 59] However, as the high risk of CVD and HF among diabetic subjects cannot be explained by an increased prevalence of atherosclerosis and hypertension alone, a specific *diabetic*

cardiomyopathy was proposed in the 1950s.[60] Experimental animal models and human studies have described multiple mechanisms, acting through the three main metabolic entities of diabetes that lead to myocardial dysfunction: hyperglycaemia, hyperinsulinaemia and hyperlipidaemia.[61] A simplified summary of these mechanisms is illustrated in Figure 2. It has been suggested that diabetic cardiomyopathy enhances hypertension-mediated cardiac and vascular damage and increases both the risk of developing HF and mortality after MI in diabetic subjects.[62]

The structural and functional changes seen in diabetic cardiomyopathy include LVH, LV diastolic dysfunction (LVDD) and LV systolic dysfunction (LVSD).[63, 64] Echocardiography, using both conventional Doppler echocardiography, and tissue Doppler imaging (TDI), is a convenient method for detecting these changes, which in many cases can be seen long before the patient develops symptoms.

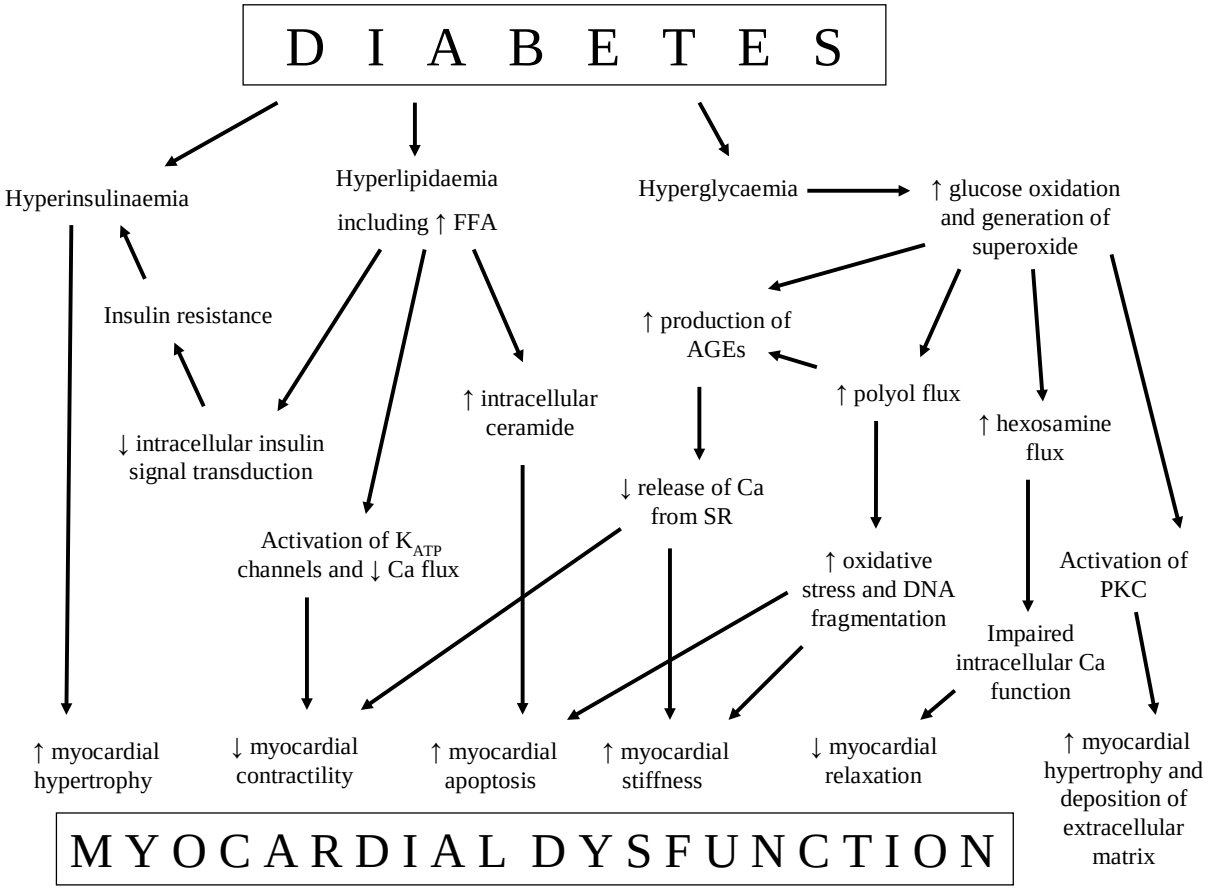

Figure 2. Mechanisms through which diabetes leads to myocardial dysfunction in ways not explained by increased prevalence of hypertension and atherosclerosis. FFA, free fatty acids; AGEs, advanced glycation end products; Ca, calcium; SR, sarcoplasmic reticulum; PKC, protein kinase C.

Screening for CVD and HF

As most of the population-attributable risks of CVD are due to modifiable risk factors, prevention should be a high priority for societies in general and health professionals in particular. Population-based prevention strategies are the most effective in the long term, but must be complemented by secondary prevention measures in high-risk individuals and subjects who already have CVD.[9, 10, 65] When identifying those at risk, risk-factor screening is unquestionably the most convenient and cheapest method, but can be complemented in selected populations by more advanced methods such as echocardiography and biomarker measurements.

Risk-factor screening

Mass screening for CVD risk factors among healthy populations has not proven to be cost-effective and is not recommended.[10, 66, 67] Screening for and treating risk factors in high-risk populations as well as secondary prevention in subjects with established CVD, are, however, effective in preventing progression of the disease and are encouraged.[10] Subjects with impaired glucose metabolism are defined as high-risk individuals. Glucometabolic impairment is often accompanied by abdominal obesity, hypertension and dyslipidaemia, as part of the metabolic syndrome, which in itself is a strong predictor of CVD.[68] Risk calculators such as the HeartScore® and the Framingham Risk Score can be useful tools to screen for and quantify CVD risk, and are widely used in both clinical and research settings.[69, 70] However, their use for subjects with impaired glucose metabolism is limited as these individuals should be considered to be at high risk irrespective of concomitant risk factors.[71] As the relationship between increasing hyperglycaemia and CVD risk is linear or J-shaped it is important to regard FPG and 2-hour plasma glucose as continuous risk factors for CVD and to take the severity of other accompanying risk factors into consideration when assessing risk.[72]

Echocardiography

Apart from risk factor screening and treatment, identifying subclinical heart disease in asymptomatic high-risk subjects is an important part of preventive cardiology. The electrocardiogram (ECG) has a relatively high sensitivity (>80%) when screening for LVSD but poor specificity limits its use.[73] Its value in detecting LVDD is also limited. However, echocardiography is probably the most useful tool for diagnosing and evaluating myocardial function. Its use is highly recommended in the current European Society of Cardiology guidelines when confirming a suspected diagnosis of myocardial dysfunction.[16]

The use of ultrasound for industrial and military purposes dates back to the early 20th century, but it was first introduced in medicine in the 1950s by two Swedish scientists, Carl Hellmuth Hertz and Inge Edler.[74] Their primary field of study was the M-mode technique, but others soon followed, introducing two-dimensional and Doppler echocardiography, which are the backbones of echocardiography today.

Two-dimensional echocardiography produces real-time images of the heart and is used for studying the morphology and dynamics of the cardiac walls, chambers and valves. The Doppler echocardiography technique relies on measurements of blood flow through the beating heart, and was initially used to evaluate valvular function. Later, methods of using pulsed-wave Doppler measurements of transmitral blood flow were developed as a means of determining LV diastolic function.[75] Passive filling of the LV in early diastole relies on the distensibility and compliance of the LV. This can be quantified by measuring transmitral blood flow in early diastole, referred to as the E-wave. The late diastolic filling phase, which occurs with left atrial contraction, is referred to as the A-wave. In LVDD, the compliance of the LV is reduced, resulting in increased LV pressure. This can affect the size and shape of the E- and A-waves. In early LVDD the E-wave is usually decreased and a compensatory increase in the A-wave is observed. However, Doppler velocities are dynamic and can vary with filling pressure at the time of measurement. The E/A pattern can also vary with degree of LVDD and age. Enlargement of the left atrium (LA) often accompanies LVDD due to increased LV pressure. The size of the LA reflects the cumulative effect of filling pressure over time and is thus regarded as a more stable indication of LVDD.[76]

Echocardiographic TDI was introduced in the 1990s.[77] TDI applies the same principles as flow Doppler echocardiography, quantifying the higher-amplitude, lower-velocity motion signals from the ventricular walls.[78] Systolic (Sm), early diastolic (Em) and late diastolic (Am) wall motion can be measured in different segments of the ventricular walls, providing an estimate of systolic and diastolic function. TDI measurements of LVDD are less load- and pressure-dependent than transmitral Doppler flow measurements and thus more stable and reliable.[79] The introduction of TDI has thus made echocardiography an even more useful imaging technique to identify patients at risk of developing HF.

The early diagnosis of cardiac abnormalities among diabetic subjects is a challenge for physicians as they often exhibit fewer symptoms of heart disease than non-diabetic subjects.[80] Indeed, echocardiographic signs of LVDD and LVH are commonly found in asymptomatic young and middle-aged diabetic subjects.[63, 81] In one study of diabetic men aged 38-67 years without known heart disease and with normal systolic function, 60% showed signs of LVDD on echocardiography.[82] IGT has also been related to asymptomatic LVDD and LVH measured by conventional echocardiography.[83-85] It has been shown that implementing treatment for asymptomatic LVH and LVSD in diabetic subjects improves survival, stressing the importance of identifying and screening subjects at risk.[80] Echocardiography can thus be regarded as an even more important and useful diagnostic tool among subjects with impaired glucose metabolism.

Nt-proBNP

As availability and cost can limit the use of echocardiography, natriuretic peptides (NPs) have been introduced as potential surrogate markers and screening tools for

LVDD and LVSD as they are relatively inexpensive and easy to use.[86] Brain or B-type natriuretic peptide (BNP), so named as it was first isolated from porcine brains in the late 1980s, is a 32-amino-acid-long polypeptide.[87] It is mainly produced by ventricular myocytes in response to volume expansion and pressure overload. BNP stimulates renal natriuresis, vasodilation, and a shift of fluids from the intravascular to the extravascular space, and suppresses the renin-angiotensin system and sympathetic activity in order to reduce the cardiac pressure overload.[88, 89] The precursor peptide, proBNP, is split into the active form (BNP) and an inactive amino-terminal called Nt-proBNP upon secretion; both of which can be measured in peripheral blood. Nt-proBNP has a longer half-life and is thus more stable but increases with the subject's age and impaired renal function to a higher degree than BNP.[89]

Both forms have been shown to sensitively identify LVSD.[73] NPs have also proven useful in the detection of LVH and LVDD.[86, 90] The use of NPs is recommended by the European Society of Cardiology for identifying patients at risk of HF, and for diagnosing and staging subjects with HF.[16] Normal values have a high negative predictive value for the presence of untreated HF, but as their concentration can be affected by multiple biological factors and diseases the positive predictive value is much lower.[91] Female sex, increasing age, impaired renal function and most diseases causing cardiac stress (CHD, valvular disease, arrhythmias, cor pulmonale, sepsis, etc.) can raise NP values, while obesity and the use of some medication can give falsely low values.[91, 92] In addition, higher NP values have been observed among subjects with diabetes, later suggested to be due to a higher prevalence of asymptomatic LV dysfunction [93, 94] and renal dysfunction[95] in diabetic subjects than in the general population. Studies have shown that BNP is a strong prognostic marker for CVD morbidity and mortality among diabetic subjects.[96, 97] However, the results of studies on the use of NPs to screen for LV dysfunction among diabetic subjects are far from consistent, making the role of NPs as a surrogate marker for LVDD and LVSD in diabetic populations uncertain.[98-104]

Self-rated health

Modern medicine often uses objective measures to estimate health and disease. CVD risk factors, echocardiographic measures of myocardial function and biomarkers such as NPs can all be categorized as such. Perceived measures of health are subjective estimates of a person's own perception of their health. Self-rated health (SRH), evaluated using a standard question*: "Would you say that in general your health is…excellent, very good, good, fair or poor?"*, has been used as a health indicator in sociological and medical research since the 1950s.[105, 106] This simple question is also a strong predictor of all-cause and disease-specific mortality among subjects with diabetes.[107] As well as being an independent risk predictor, poor SRH has been shown to strengthen the predictive value of biomedical risk factors for MI and stroke.[108, 109] There are, however, many factors other than chronic physical illness that influence SRH ratings, the most important being social and mental well-being, and these can vary with age, gender and cultural norms in different societies.[110] This instrument thus

gives rather a crude measure of health and therefore has limited application in the clinical setting, but it is often used in epidemiological research because of simplicity.

Gender differences in diabetic heart disease

Although the excess relative risk of CVD among women with diabetes compared to men with diabetes seems to have declined since it was first described in the Framingham Heart study in Massachusetts, USA in the 1970s, recent studies still indicate that women bear a disproportionate part of the diabetes-related excess risk. A recent meta-analysis has shown that the relative risk of fatal CHD in diabetic compared to non-diabetic subjects is 3.5 (95% confidence interval (CI) 2.7-4.5) for women and 2.1 (95% CI 1.81-2.34) for men.[111] While the lifetime risk and mortality rates for CVD in the general population are similar for men and women, there are pathophysiological, epidemiological, prevention- and treatment-related differences in CVD between the sexes (Table 1).[112] These probably play a role in the relative discrepancy in risk for CVD incidence and mortality between diabetic men and women. Some specific explanations have been highlighted in the literature. Studies have shown a heavier risk factor burden for diabetic women than diabetic men.[113, 114] The Strong Heart Study, a US study which included American Indians, showed greater differences in women with diabetes than in those without diabetes for waist-to-hip ratio, HDL, apoB, apoA1, fibrinogen and low-density lipoprotein (LDL) size, compared to men.[115] Also, the relative impact of concomitant risk factors, especially hypertension and dyslipidaemia, seems to be greater for diabetic women than for diabetic men.[116] Some authors have suggested that diabetes in itself is a more potent risk factor in women.[6, 116] Disparities in diagnosis and medical treatment of heart disease between diabetic men and women have also been described.[117] Various biological explanations have been suggested including a higher pro-thrombotic state and increased oxidative stress in diabetic women than in diabetic men, adverse effects of sex hormones and differences in endothelial function between the sexes.[118-120]

Ageing populations

Due to an increase in the ageing population and the associated increase in the prevalence of impaired glucose metabolism, CVD and HF, elderly subjects constitute a large part of the patient population affected by these conditions. At the same time, most studies from which current evidence is gathered are based on populations comprising young and middle-aged subjects, often with male predominance.

Although not as well documented as in younger cohorts, impaired glucose metabolism also predicts future CVD and HF amongst the elderly.[121, 122] However, it has been shown that risk ratios for the incidence of CVD and HF between diabetic and non-diabetic subjects decrease with age.[123, 124]

Table 1. Gender differences in cardiovascular disease.

Subject	Important aspects in women
Epidemiology	7-10 years later onset of CVD More disability-adjusted life years lost to CVD at older age
Atherosclerosis	Differences in inflammation and oxidative stress Lower atheroma burden at younger ages (<65 years) Oestrogens involved in plaque composition/vascular function Vascular dysfunction and small-vessel disease ACS with "normal" or non-obstructive coronary artery disease More plaque erosions than plaque ruptures at ACS
Heart failure	Hypertension and diabetes main causes of heart failure Predominant heart failure with preserved LV ejection fraction Elderly women have more LV hypertrophy (men more fibrosis)
Thrombosis	Changes in platelet activity, coagulation factors and fibrinolytic activity related to hormone status pre-/post-menopause, pregnancy, etc. More bleeding complications after interventions Increased risk of thrombosis with atrial fibrillation
Risk factors	Hypertension Higher prevalence at older age Higher association with strokes, LV hypertrophy and diastolic HF Diabetes >50% higher CVD mortality Diffuse atherosclerosis, higher co-morbidity Independent risk factor for HF Lipids Low HDL and elevated TG more related to CVD
Sex-related risk factors	Pregnancy-related hypertension and gestational diabetes Hormonal dysfunction pre-menopause/PCO Menopause
Lifestyle/ psychosocial factors	Smoking <55 years gives higher risk of ACS Obesity/physical inactivity Anxiety/stress and lower socio-economic status
Diagnosis	Differences in symptom presentation/communication More angina with less obstructive coronary artery disease Lower sensitivity and specificity of non-invasive testing
Therapy	Gender differences in efficacy/interaction/side effects

Adapted from Maas et al, Red alert for women's heart: the urgent need for more research and knowledge on cardiovascular disease in women. *Eur Heart J* 2011:32(11); page 1368. By permission of Oxford University Press. ACS, acute coronary syndromes; PCO, poly-cystic ovary disease.

Few studies have been published on the relationship between glucose metabolism and LV structure and function measured with echocardiography in otherwise healthy elderly subjects, and these have revealed conflicting results.[125-127] The same applies to NPs; most studies on the use of NPs to detect asymptomatic LV dysfunction among subjects with impaired glucose metabolism have focused on younger and middle-aged subjects with established diabetes.[93, 94, 128] Studies in elderly and pre-diabetic subjects are sparse. Additionally, gender-specific analyses are lacking in all the above-mentioned areas of research.

In light of the enormous personal disability and the cost to society of impaired glucose metabolism, CVD and HF it is important to establish whether the same screening strategies and diagnostic methods can be applied to elderly subjects with impaired glucose metabolism as to younger and middle-aged subjects, and the degree to which the relationships between impaired glucose metabolism, CVD risk factors and CVD and HF morbidity and mortality are affected by age and ageing.

In summary, as the world's population is becoming older the prevalence of impaired glucose metabolism, CVD and HF will increase correspondingly. Most of the population-attributable risks for CVD are modifiable. Prevention should thus be a high priority, especially in high-risk populations such as subjects with diabetes, of which almost 70% succumb to CVD. Apart from risk factor screening and treatment, identifying subclinical heart disease is important. The method of choice for identifying subclinical LV dysfunction is echocardiography, applying both traditional Doppler echocardiography and TDI. On account of cost and limited access to echocardiography, NPs have been used as surrogate markers for LV dysfunction, especially in the primary care setting. The role of NPs as screening tools among subjects with diabetes has, however, been questioned. Also, as incidence ratios for CVD and HF between diabetic and non-diabetic subjects seems to fall with age the role of screening for subclinical heart disease among elderly subjects with impaired glucose metabolism is unclear. As the majority of those suffering from impaired glucose metabolism, CVD and HF are elderly it is important to study the effects of age, in order to improve strategies for prevention and treatment.

The *overall aim* of the work presented in this thesis was to study gender-related associations between glucose metabolism and myocardial dysfunction (by means of echocardiography and Nt-proBNP), risk factor burden and CVD morbidity and mortality in middle-aged and elderly subjects. The hypothesis tested was that similar associations would be observed as previously seen in younger populations, although a weakening of the associations might be expected with advancing age.

AIMS

The aims of the work presented in this thesis are given below.

- To examine whether glucose metabolism in groups classified by increasing glucometabolic impairment was associated with measures of LV diastolic function and LVH, evaluated by conventional Doppler echocardiography and TDI in two independent population-based cohorts of middle-aged and elderly men and women (Paper I).

- To study the associations between Nt-proBNP and FPG as a continuous variable and by grouping in a population-based cohort, grouped according to gender and age into middle-aged and elderly subjects (Paper II).

- To compare the strength of the correlation between glucose metabolism (in groups defined by increasing glucometabolic impairment) and: 1) CVD risk factor burden and 2) SRH between middle-aged and elderly men and women (Paper III).

- To evaluate the influence of age on the association between glucose metabolism (in groups defined by increasing glucometabolic impairment, ranging from normal fasting glucose to established diabetes) and CVD event risk among middle-aged and elderly men and women (Paper IV).

STUDY POPULATIONS AND METHODS

Study subjects

The Malmö Preventive Project

The Malmö Preventive Project (MPP) was a large-scale population-based cohort study conducted in Malmö, southern Sweden between 1974 and 1992. The project was a preventive case-finding programme with the aim of screening for CVD risk factors, alcohol abuse and breast cancer in the general population.[129] A flow chart explaining the recruitment is shown in Figure 3. In short, birth cohorts of inhabitants of Malmö, the third largest city in Sweden, were invited to participate in the project (men born in the years 1921, 1926-1942, 1944, 1946 and 1948-9; women born in 1926, 1928, 1930-6, 1938, 1941-2 and 1949); 33,346 young adults and middle-aged individuals participated.[66, 130]

Re-screening of the MPP participants, the *MPP Re-Examination Study (MPP-RES)*, was conducted at Skåne University Hospital between 2002 and 2006. The target population consisted of the approximately 25,000 individuals from the original MPP cohort, still alive and living in the Malmö area. In total, 18,238 middle-aged and elderly subjects participated (mean age 69±6 years, range 57-86 years) (Figure 3). After receiving verbal information about the study, blood samples were drawn from each individual (after overnight fasting). Whole blood was stored in a biobank for later genetic analysis.[131] At the second visit blood pressure and pulse rate were measured twice in the supine position after 5 minutes' rest. Height, weight, waist and hip circumferences were measured in light indoor clothing without shoes. The participants then answered a self-administered questionnaire on lifestyle (smoking, alcohol consumption and physical activity), stress, their general state of health, short medical history, and medication.

Echocardiography was performed and a resting ECG obtained in a subsample of 1,792 participants on a separate occasion. The subjects were randomly selected from groups defined by glucose metabolism: normal fasting glucose (NFG, ≤6.0 mmol/l); IFG (according to the WHO definition); new-onset type 2 diabetes; and established diabetes (type 1 or type 2); with oversampling in groups of subjects with impaired glucose metabolism to ensure sufficient numbers of subjects studied in each group.

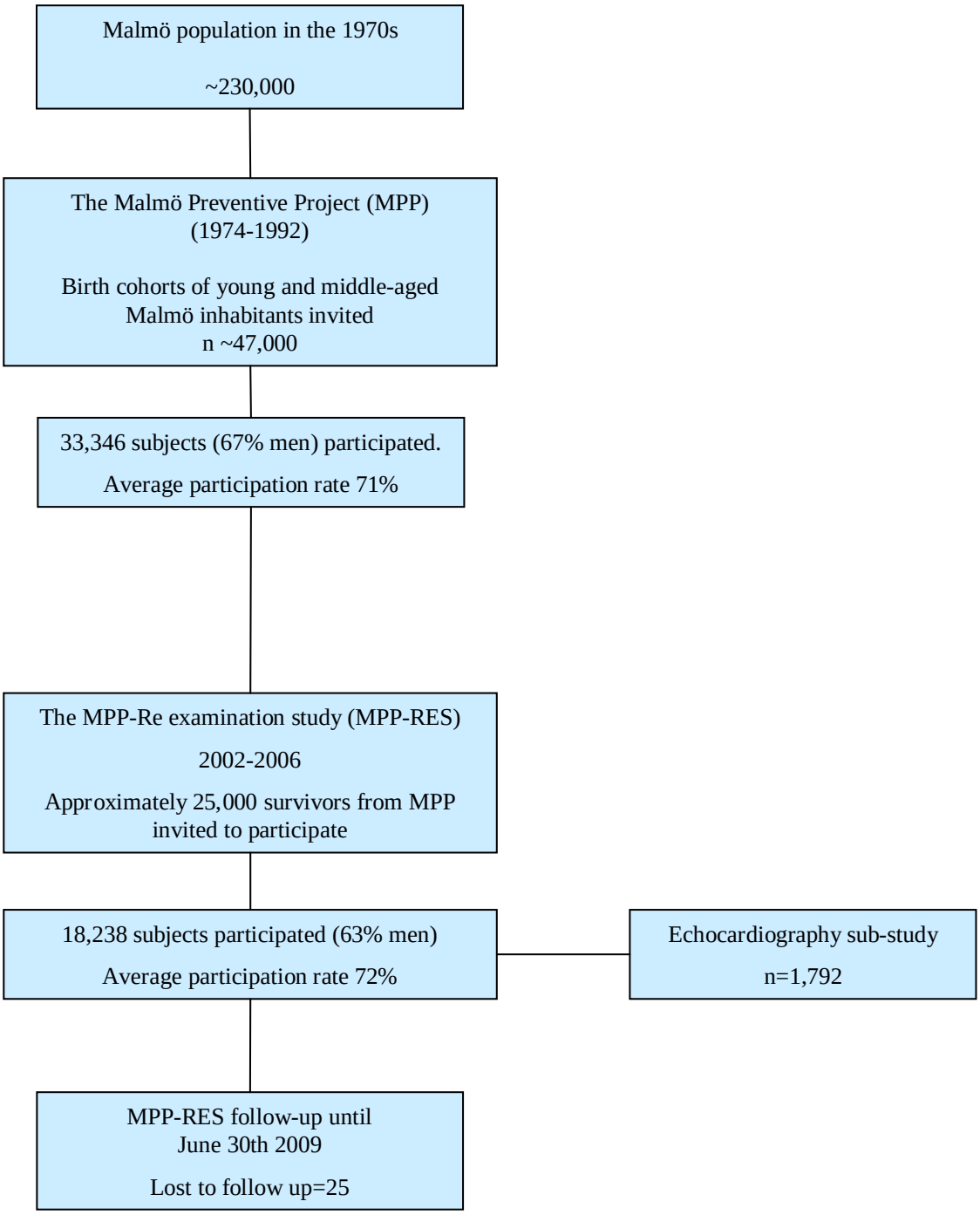

Figure 3. Schematic description of recruitment in the MPP and MPP-RES studies.

The AGES-Reykjavik Study

The Reykjavik Study (1967-1997) was a prospective cohort study, designed to study the epidemiology of CVD risk factors in the Icelandic population.[132] Approximately 19,400 inhabitants, born during the period 1907-1935, living in the greater Reykjavik area on 1st December 1966, participated in the study.

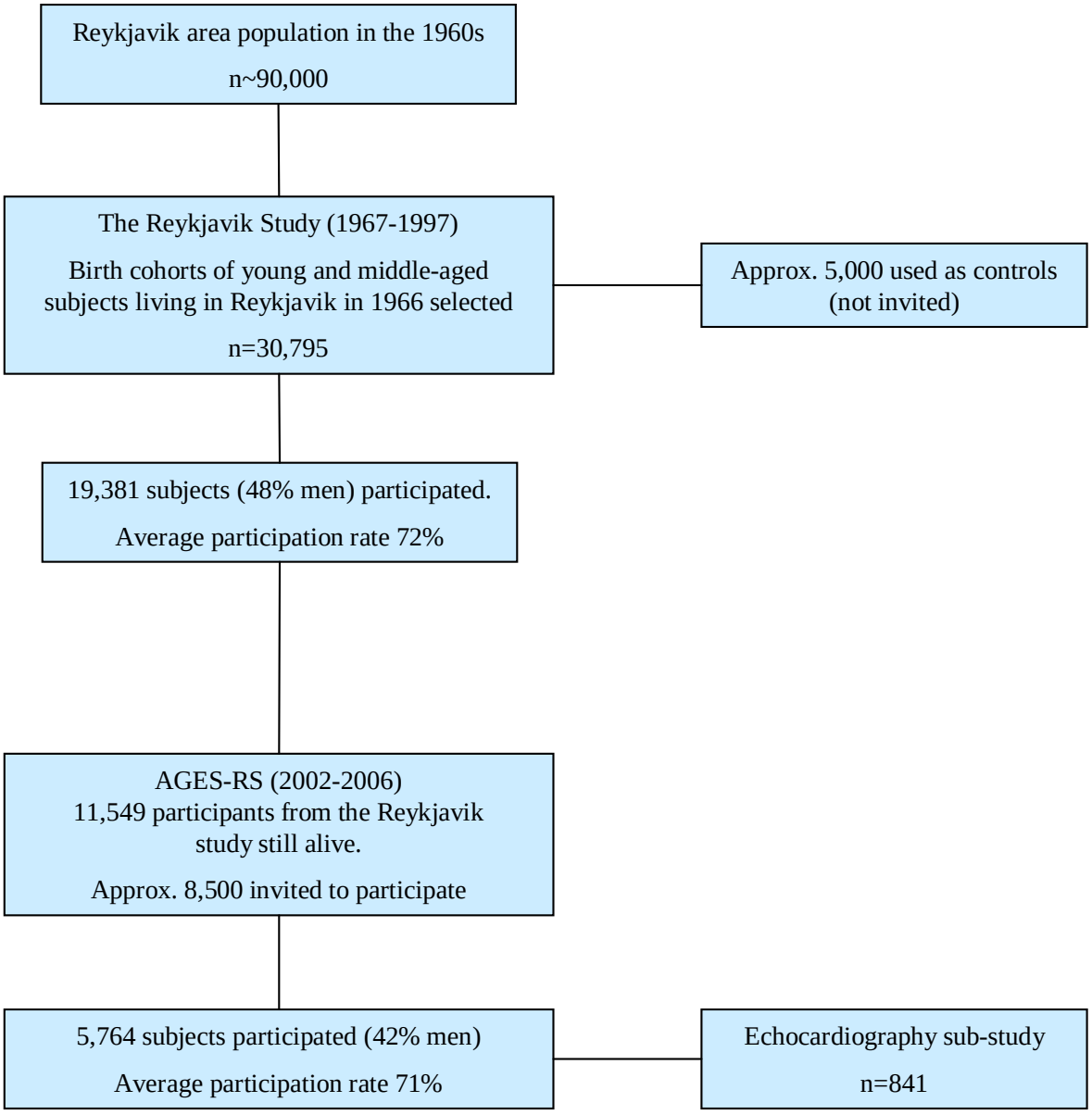

Figure 4. Schematic description of the Reykjavik Study and the AGES-RS.

The Age Gene/Environment Susceptibility Reykjavik Study (AGES-RS), conducted in 2002-2006, was a prospective study based on the original Reykjavik Study cohort, designed to examine risk factors in relation to disease and disability in old age.[133] In total, 5,764 mostly elderly subjects attended the study (mean age 76±6 years, range 67-95 years). The examination included an extensive questionnaire and the collection of a blood sample. An ECG was recorded and blood pressure, heart rate (two separate measurements), height and weight were measured. An echocardiography study was performed on a subsample of 841 randomly selected individuals. A schematic description of the participation protocol for both studies can be seen in Figure 4.

Echocardiography

The echocardiography studies were conducted with a 3V2c transducer (Acuson Sequoia, Mountain View, CA, USA; AGES-RS and MPP-RES) or an S3 transducer (Sonos 5500 Philips, Andover, MA, USA; MPP-RES). All subjects were studied in the left lateral decubitus position. Parasternal long- and short-axis, and apical four- and two-chamber views were used to evaluate cardiac dimensions and the left ventricular ejection fraction (LVEF).[134] LV diastolic function was measured using transmitral pulsed Doppler flow as well as TDI in the four-chamber view.[135] Transmitral Doppler measurements were conducted with the pulsed wave sample volume between the tips of the mitral valve leaflets in the early (E) and late (A) diastolic phases. TDI was performed with the sample volume close to the mitral annulus in the lateral and septal LV walls in the apical four-chamber view. Sm, Em and Am were measured. In the MPP-RES, a single cycle was used to calculate the transmitral and TDI parameters if the recordings were homogeneous; otherwise a mean of three to five cycles was used, as was done in the AGES-RS for all calculations. LV mass calculations were based on two-dimensional end-diastolic measurements in the parasternal long-axis view at the level of the mitral tips.[134] In both studies LVEF was quantified visually.

In the AGES-RS, the inter-observer variability between the three readers performing the studies and a reference reader from the US National Institute of Health was investigated on selected variables. Intra-observer variability was not assessed in the AGES-RS. Intra- and inter-observer variability in the MPP-RES was tested by two readers independently analysing images from a random sub sample of subjects (Table 2).

Laboratory tests

The laboratory tests carried out on the MPP-RES cohort included FPG and serum (s-) lipid profile[a]. In the echocardiography subcohort Nt-proBNP[b], plasma cystatin C[c] and HbA1c[d] (MonoS standard) were measured. For logistic reasons HbA1c measurements were only performed in 60% of the subjects, randomly selected. All measurements were carried out at the Department of Clinical Chemistry, Skåne University Hospital, Malmö, apart from Nt-proBNP which was measured at the Department of Clinical Chemistry, Akershus University Hospital, Lorenskog, Norway. In the AGES-RS

[a] Beckman Coulter LX20, Beckman Coulter Inc., Brea, CA, USA
[b] Electrochemiluminescence immunoassay, Elecsys, Roche Diagnostics, Basel, Switzerland
[c] Automated particle-enhanced immune-turbidimetric method with reagents from DakoCytomation, Glostrup, Denmark
[d] Automated HPLC, Variant II from Bio-Rad Laboratories, Munich, Germany

cohort, s-fasting glucose (FG), HbA1c (DCCT standard), s-insulin, s-creatinine and s-lipid profile were measured[e] at the local Reykjavik Study laboratory.

Table 2. Coefficient of variation (%) for inter- and intra-observer variability in the MPP-RES and AGES-RS cohorts.

	IVSd	LVPWd	LVIDd	E	A	Em	Am
MPP-RES							
Inter-observer variability	13.0	12.1	4.1	2.6	2.4	4.4	6.6
Intra-observer variability	10.5	5.5	3.3	1.1	1.2	2.8	3.7
AGES-RS							
Inter-observer variability	13.2	9.1	5.1	4.0	3.5	5.2	3.9
Intra-observer variability	NA	NA	NA	NA	NA	NA	NA

IVSd, interventricular septum diameter at end-diastole. LVPWd, left ventricular posterior wall diameter at end-diastole. LVIDd, left ventricular internal diameter at end-diastole. E, peak velocity of the transmitral E-wave. A, peak velocity of the transmitral A-wave. Em, early diastolic peak velocity in septal LV wall. Am, late diastolic peak velocity in septal LV wall. NA, not assessed.

Description of variables

Impaired glucose metabolism

Impaired fasting glucose was defined according to the WHO criteria (FG 6.1-6.9 mmol/l) in both the MPP-RES and AGES-RS cohorts.[29] In the MPP-RES cohort, new-onset type 2 diabetes was defined by two separate FPG values $\geq$7.0 mmol/l,[29] or a single measurement $\geq$11.1 mmol/l. Those with one measurement of FPG 7.0-11.0 mmol/l and the other $\leq$6.9 mmol/l were grouped with the IFG subjects. This was done to maximize the benefit of having two separate measurements and thus minimize the risk of misclassification. In the AGES-RS cohort, s-FG was only measured once. Thus, a single s-FG $\geq$7.0 mmol/l was defined as new-onset type 2 diabetes. In both the MPP-RES and the AGES-RS cohorts, individuals with a history of type 1 or type 2 diabetes (according to the questionnaire or medication) were classified as having established diabetes regardless of their glucose level. No OGTTs were performed in these studies.

[e] Elecsys 1010 (s-insulin) and Hitachi 912 (s-FG, HbA1c, s-creatinine and lipid profile), Roche Diagnostics, Basel, Switzerland

Incident and prevalent heart disease

In Paper I, a self-reported diagnosis of HF was used to define prevalent HF in both cohorts. In addition, in the MPP-RES cohort a diagnosis of HF by means of International Classification of Diseases (ICD) codes acquired from the local hospital diagnosis registry was used to define prevalent HF (ICD-10 code I50). Similarly, MI diagnoses retrieved from the local hospital diagnosis registry (ICD-10 codes I21, I22 and I25.2) were used to define prevalent CHD. In both cohorts, prevalent valvular disease was defined as 1) aortic stenosis with maximum transvalvular Doppler flow velocity ≥3.0 m/s, 2) severe aortic-, 3) mitral- or 4) tricuspid regurgitation, assessed by echocardiography.[136]

In Paper II prevalent CHD and significant valvular disease were defined in the same way as in the MPP-RES cohort in Paper I. In addition to the self-reported diagnosis of HF, a LVEF≤40% at the tim e of the echocardiography was classified as prevalent HF. Also, patients answering "no" to having HF but having a previous ICD diagnosis of HF, LVEF 41-55% and who were taking two or more prescription drugs for the treatment of HF, were also classified as having prevalent HF.

In Paper IV incident CVD events (coronary and cerebral) as well as prevalent CVD and HF were defined by ICD-9 and ICD-10 codes gathered from the Swedish Hospital Discharge Registry and the Cause-of-Death Registry from the Centre for Epidemiology at The Swedish National Board of Health and Welfare. Additionally, stroke events were extracted from local hospital and study registries. Incident CVD events included the following (ICD-9 codes in brackets): acute MI (410), other acute and sub-acute forms of ischaemic heart disease (411), old MI (412), angina pectoris (413), other forms of chronic ischaemic heart disease (414), occlusion of cerebral arteries (434), subarachnoidal haemorrhage (430), intra-cerebral haemorrhage (431) and acute but ill-defined cerebrovascular disease (436). Prevalent CVD included the same diagnoses. Prevalent HF included heart failure (428) and hypertensive heart disease with heart failure (402.01, 402.11 and 402.91).

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CVD risk factors

Uncontrolled hypertension was classified as present if subjects had systolic blood pressure (SBP) ≥140 mmHg and/or diastolic blood pressure (DBP) ≥90 mmHg.
Body mass index (BMI, kg/m^2) was calculated from weight (kg) and height (m).
Central obesity was classified as present if subjects had a waist circumference of ≥102 cm for men and ≥88 cm for women.
Dyslipidaemia was classified as present if the subjects had a total cholesterol level of ≥5.0 mmol/l, LDL ≥3.0, TG ≥1.7 mmol/l and/or HDL <1.3 mmol/l for men and 1.0 mmol/l for women. LDL was calculated using the Friedewald's formula.[137]
Limits for blood pressure, waist circumference and lipids were based on the European Society of Cardiology guidelines for CVD prevention (blood pressure, total cholesterol, LDL) and classification of the metabolic syndrome (TG, HDL, waist circumference).[10, 21]

Physical activity was self-reported in the questionnaire: *"How physically active are you today? 1) Physically inactive in my spare time 2) Limited physical activity in my spare time 3) Regular moderate physical activity 4) Regular heavy physical activity or competitive sports"*. Those reporting no physical activity were classified as being physically inactive.
Smoking was self-reported, by the question *"Do you smoke? 1) Yes 2) No (previous smoker) 3) No (never smoked)"*.

Medication for CVD or hypertension was defined as calcium-channel blockers, beta-receptor blockers, angiotensin-converting enzyme inhibitors, angiotensin-receptor blockers, long-acting nitrates, anti-platelet agents, diuretics, and lipid-lowering drugs.

SRH was measured in the questionnaire using the standardized question "*Would you say that in general your health is…Excellent, Very good, Good, Fair or Poor?*"[105] Subjects were classified as having low SRH when answering fair or poor.

Study protocols and statistical analysis

In all four studies included in this thesis, the subjects were divided into six groups defined by FG and impaired glucose metabolism: 1) FG ≤5.0 mmol/l; 2) FG 5.1 -5.5 mmol/l; 3) FG 5.6-6.0 mmol/l; 4) IFG (FG 6.1-6.9 mmol/l); 5) new-onset type 2 diabetes and; 6) established diabetes.

Baseline characteristics are presented as means ± 1 standard deviation (SD) and percentages. Variables with a skewed distribution were naturally log-transformed. The common significance level of $p<0.05$ (two-tailed) was used. All calculations were performed using SPSS 16.0-19.0 (SPSS Inc., IL, USA).

Paper I

The data from the MPP-RES and AGES-RS echocardiography subcohorts were used in Paper I and were analysed separately. Data were first analysed for both sexes and then divided according to gender. In a post hoc analysis (using the MPP-RES cohort only), the calculations were repeated after dividing the subjects into two groups: middle-aged (57-69 years) and elderly (70-80 years). The study was of cross-sectional design.

Subjects with prevalent CHD, HF and significant valvular disease were excluded from all calculations. Data from the glucometabolic groups were entered into a multivariable regression analysis and tested against measures of LV diastolic function and structure, using the first group (FG ≤5.0 mmol/l) as reference group. The endpoint variables and their definitions are listed in Table 3.

Table 3. Evaluated measures of LV function and structure and their definitions.

Variable	Definition
Em	Peak myocardial velocity of the basal left ventricular lateral wall in early diastole (relaxation of the left ventricle accompanying passing filling) measured with TDI
Am	Peak myocardial velocity of the basal left ventricular lateral wall in late diastole (relaxation accompanying active filling of the left ventricle during atrial systole) measured with TDI
Em/Am	The ratio between Em and Am
Transmitral E	Doppler measurement of peak velocity of blood flow through the mitral valve in early diastole (passive filling of the left ventricle)
Transmitral A	Doppler measurement of peak velocity of blood flow through the mitral valve in late diastole (active filling of the left ventricle during atrial systole)
Transmitral E/A	The ratio between transmitral E and transmitral A
Transm. E/Em	The ratio between transmitral E and Em
LAD	Left atrial diameter at ventricular end-systole
LVMI	Left ventricular mass [a] indexed for body surface area (a quantitative measure of left ventricular hypertrophy)[a]

[a] According to the formula: $0.8*(1.04*((IVSd+LVIDd+LVPWd)^3-LVIDd^3))+0.6$.[134]
IVSd, interventricular septum diameter; LVDd, left ventricular internal diameter; LVPWD, left ventricular posterior wall diameter; all measured at end-diastole.

The regression analyses were first age-adjusted and subsequently fully adjusted. In order to select relevant covariates, the relationship between each covariate and LV function/structure variable was tested by regression analysis. Covariates showing a strongly significant correlation with the relevant LV function/structure variable ($p<0.01$) were then tested for internal correlation (using Spearman's rank test or Pearson's test). If internal correlation was present ($r\geq0.3$), only the covariate with the strongest correlation to the relevant LV function/structure variable was incorporated into the final multivariable analysis. The covariates tested against each variable are listed in Table 4.

The associations between FG (FPG in the MPP-RES cohort, s-FG in the AGES-RS cohort), HbA1c and an estimate of insulin resistance using the homeostasis model assessment (HOMA: s-insulin*FG/22.5; AGES-RS only)[138] and the different measures of LV diastolic function and LVMI were then tested using the same model as described above. Patients with established diabetes were excluded from these calculations as one can expect variations in FG, HbA1c and HOMA values in these subjects depending on their treatment. Also, better treatment of subjects with diabetes could influence the echocardiographic findings, resulting in non-linearity.

Table 4. Covariates tested against each endpoint variable (Paper I).

Covariate
Age
Gender
BMI
Smoking
s-lipids
Measures of renal function (s-creatinine in the AGES-RS cohort, cystatin C in the MPP-RES cohort)
SBP and DBP
Heart rate
LVMI (only against LV function variables)
Medication for CVD and hypertension
Atrial fibrillation or flutter

Paper II

All calculations described in Paper II were performed on data from the MPP-RES echocardiography subcohort divided according to gender and age, using the same age grouping as in the post-hoc analysis in Paper I. The study was of cross-sectional design.

The association between Nt-proBNP and FPG was tested by linear regression analysis with backwards-stepwise selection, in several steps, as shown in Table 5. Calculations were repeated after excluding subjects with established diabetes, prevalent CHD, HF or significant valvular disease.

The mean values of Nt-proBNP in the six glucometabolic groups were then calculated and compared using a univariate general linear regression model, adjusting for all the covariates listed in Table 5, with and without subjects with prevalent CHD, HF or significant valvular disease.

In a post hoc analysis, the degree of explanation (R^2) for the variance in Nt-proBNP was assessed using linear regression, including all the variables in the previous models as well as complementary echocardiography variables reflecting LV diastolic function (transmitral E/Em ratio and LA area adjusted for body surface area).

Table 5. Adjustment in the linear regression analysis presented in Paper II.

Step
1. Unadjusted
2. Adjusted for age
3. Age and cystatin C
4. Age, cystatin C and BMI
5. Age, cystatin C, BMI, uncontrolled hypertension, heart rate, HDL, TG, smoking and CVD disease/hypertension medication.
6. All covariates in 5. plus atrial fibrillation or flutter, LVMI and echocardiographic measures of LV systolic and diastolic function.[a]

[a] LVEF, Sm, Em and Am were chosen as variables reflecting LV systolic and diastolic function because they showed the strongest correlation with FPG in the first study (Paper I).

Paper III

In Paper III, which was also of cross-sectional design, the whole MPP-RES cohort (n=18,238) was used. All calculations were performed on the data grouped according to gender and age.

The proportions of MPP-RES subjects (%) having: a) at least three of five chosen CVD risk factors (uncontrolled hypertension, dyslipidaemia, central obesity, current smoking or lack of PA) and b) low self-rated health (SRH) were calculated for each of the six glucometabolic groups. Differences in proportions between age groups within each glucometabolic group were calculated using simple (unadjusted) logistic regression, generating odds ratios (ORs) and 95% confidence intervals (95% CI). To assess the strength of the correlation between glucometabolic impairment and risk factor burden and poorer SRH, logistic regression analysis was used to calculate beta (β)-coefficients for trends from the first to the sixth group for SRH and for the first to the fifth group for the risk factors, excluding the sixth group (those with established diabetes) for the same reasons as in Paper I. Ratios between the β-coefficients obtained from the trend analyses were then calculated, and interaction analysis was performed to estimate differences between the age groups, by entering variable A *variable B into the regression analysis.

Paper IV

Data from the whole MPP-RES cohort were used in the study described in Paper IV, which was of prospective cohort study design. As in the previous studies, the data were analysed in gender and age groups.

Cox's proportional hazard regression analysis was used to estimate hazard ratios (HRs) and 95% CI for fatal or non-fatal CVD events (coronary or cerebral) for the different glucometabolic groups, using the first group (FPG ≤5.0 mmol/l) as a reference group. Also, trends from the first to the fifth group were explored (excluding subjects with established diabetes), by entering the grouping variable as a continuous variable into the Cox model. Interaction analysis was applied to explore differences in trends between the two age groups and between men and women. Follow-up time was from the date of entry into the study until the end of follow-up (30th June 2009), death, "lost to follow-up" or the occurrence of an endpoint. Adjustments were made for confounding by DBP, LDL, HDL, TG, waist circumference, current smoking, physical inactivity, prevalent CVD or HF at baseline, as well as medication for CVD and hypertension. The correlation between SBP and DBP was strong ($r=0.74$, $p<0.0001$) thus only DBP was included in the regression model, as it had a stronger correlation to the endpoints than SBP. The same was true for BMI vs. waist circumference ($r=0.79$, $p<0.0001$), and total cholesterol vs. LDL ($r=0.93$, $p<0.0001$), where the latter variable in both cases showed a stronger correlation to the endpoints. Current smoking, physical inactivity and medication were self-reported in the questionnaire and were entered into the regression model as dichotomous variables.

Ethical considerations

All participants in both the MPP-RES and the AGES-RS signed an informed consent form before entering the studies. Participants in the MPP-RES were given verbal and written information on the laboratory tests and blood pressure measurements, and were offered an appointment with a physician if new-onset type 2 diabetes, hyperlipidaemia and/or hypertension were detected, if they did not have regular contact with their family physician. The Ethics Committee of Lund University, Sweden, approved the original MPP (Official records no. 85/2004) and MPP-RES (LU 244-02). The National Bioethics Committee in Iceland and the Icelandic Data Protection Committee approved AGES-RS. Both studies complied with the Declaration of Helsinki.

RESULTS

Baseline characteristics

The baseline characteristics of the men and women in the echocardiography subcohorts (Papers I-II), are given in Tables 6 (MPP-RES, n=1,792) and 7 (AGES-RS, n=841). As age stratification was only applied to the MPP-RES cohort, the baseline characteristics for the AGES-RS echocardiography subcohort are presented without age stratification. The first three glucometabolic groups are combined in the tables for simplification as these subjects all had NFG. The baseline characteristics for the whole MPP-RES cohort (n=18,238), divided into two age groups can be seen in Table 8 (Papers III-IV). As in the previous tables the first three glucometabolic groups were combined. FPG values were not available for 12 non-diabetic subjects and could thus not be categorized.

Paper I

The AGES-RS cohort

Table 9 gives raw mean values (± 1 SD) of each LV function/structure variable for the six glucometabolic groups and adjusted p-values from the multivariable analysis for the intergroup trend tests (groups 1-5). Subjects with prevalent CHD, HF and significant valvular disease (n=233) were excluded from all calculations. No significant differences were observed between the glucometabolic groups, nor were any of the trend tests statistically significant.

The associations between s-FG, HbA1c and HOMA and LV function/structure variables are given in Table 10. HbA1c was significantly inversely correlated with Am. After applying age-adjustment, HOMA was positively correlated with transmitral E, transmitral A and the E/Em ratio. LVMI was positively significantly associated with s-FG (age-adjusted) and HbA1c (fully adjusted).

When data from the men and women were analysed separately, the correlations between HOMA and transmitral A were stronger in men (age-adjusted *F*-statistic 11.5; p=0.001) than in women (age-adjusted *F*-statistic 4.0; p=0.048), while correlations between HOMA and transmitral E (age-adjusted *F*-statistic 7.6; p=0.006) and E/Em (age-adjusted *F*-statistic 11.2; p=0.001; fully adjusted *F*-statistic 6.4; p=0.01) were only seen in women. Among men there was a significant trend towards higher Em with greater impairment of glucose metabolism (by groups; fully adjusted p-trend=0.008), a positive correlation between s-FG and Em (fully adjusted *F*-statistic 5.3; p=0.02), a negative correlation between HOMA and Em/Am ratio (age-adjusted *F*-statistic 12.6; p=0.001; fully adjusted *F*-statistic 10.0; p=0.002) and transmitral E/A (age-adjusted *F*-statistic 8.4; p=0.004; fully adjusted *F*-statistic 7.3; p=0.008).

Table 6. Baseline characteristics of the MPP-RES echocardiography subcohort (Papers I-II).

	Middle-aged (57-69 years)				Elderly (70-79 years)			
MEN	**NFG**	**IFG**	**New-onset diabetes**	**Established diabetes**	**NFG**	**IFG**	**New-onset diabetes**	**Established diabetes**
Number (n)	265	302	112	193	120	96	33	145
Mean age (years)	63±4	63±4	63±3	64±4	74±3	74±2	74±2	74±2
SBP (mmHg)	142±17	150±19	157±21	147±19	145±19	152±22	148±25	147±20
DBP (mmHg)	84±10	88±10	90±10	84±10	83±10	84±10	82±9	81±11
BMI (kg/m^2)	27±3	28±4	30±5	29±4	26±3	28±3	29±4	29±4
Total cholesterol (mmol/l)	5.6±0.9	5.6±1.1	5.7±1.2	4.9±1.1	5.3±1.1	5.1±1.1	5.0±0.9	4.7±1.0
LDL (mmol/l)	3.0±1.1	3.1±1.1	3.5±1.2	2.8±1.1	2.8±1.1	2.7±1.1	2.7±1.1	2.5±0.9
HDL (mmol/l)	1.3±0.3	1.2±0.3	1.1±0.3	1.1±0.3	1.3±0.4	1.3±0.3	1.2±0.3	1.2±0.3
TG (mmol/l)	1.2±0.7	1.5±1.0	2.2±1.8	1.7±1.2	1.2±0.6	1.4±0.7	1.7±0.9	1.6±0.8
FPG (mmol/l)	5.5±0.4	6.5±0.4	8.9±2.7	8.6±2.7	5.3±0.4	6.5±0.5	8.7±2.0	8.7±2.4
HbA1c (%)[a]	4.5±0.3	4.8±0.4	5.7±1.1	6.4±1.4	4.7±0.3	4.9±0.4	5.9±0.6	6.7±1.5
Nt-proBNP (pmol/l)	14±27	15±29	20±43	20±34	45±77	49±68	85±166	54±137
Cystatin C (mg/l)	1.1±0.2	1.0±0.2	1.0±0.2	1.1±0.4	1.2±0.2	1.2±0.2	1.2±0.3	1.3±0.4
Current smoker (%)	25	23	24	17	17	19	21	15
Using medication for CVD or HTN (%)	37	41	46	76	58	73	79	87

[a] By Mono-S standard. Only measured in approximately 60% of the subjects.

Table 6, continued.

	Middle-aged (57-69 years)				Elderly (70-80 years)			
WOMEN	**NFG**	**IFG**	**New-onset diabetes**	**Established diabetes**	**NFG**	**IFG**	**New-onset diabetes**	**Established diabetes**
Number (n)	126	53	14	38	133	61	23	78
Mean age (years)	65±4	65±4	66±3	66±4	73±2	73±2	74±3	73±3
SBP (mmHg)	142±19	142±16	148±22	143±22	144±19	147±22	158±27	147±18
DBP (mmHg)	83±10	83±8	84±9	80±9	82±10	84±12	87±11	81±9
BMI (kg/m^2)	27±4	30±5	33±7	32±6	27±4	30±5	31±6	29±5
Total cholesterol (mmol/l)	6.0±1.0	5.8±1.1	6.3±1.3	5.2±1.0	5.9±1.0	5.8±1.2	5.8±0.9	5.2±0.9
LDL (mmol/l)	2.7±0.9	2.7±1.2	3.4±1.1	2.6±1.1	2.4±0.9	2.6±1.0	3.1±0.9	2.2±0.8
HDL (mmol/l)	1.5±0.4	1.5±0.5	1.2±0.3	1.3±0.5	1.7±0.5	1.5±0.5	1.3±0.4	1.5±0.4
TG (mmol/l)	1.2±0.7	1.4±0.7	2.1±0.6	1.5±0.8	1.2±0.6	1.5±0.6	1.6±0.6	1.4±0.7
FPG (mmol/l)	5.3±0.4	6.4±0.4	11.2±4.5	8.2±2.2	5.2±0.4	6.5±0.6	8.1±1.0	8.1±2.1
HbA1c (%)[a]	4.6±0.4	4.9±0.4	5.5±0.5	6.7±1.4	4.7±0.3	5.0±0.5	5.6±0.3	6.4±0.8
Nt-proBNP (pmol/l)	21±54	21±39	14±11	20±43	31±37	28±30	33±54	44±68
Cystatin C (mg/l)	1.1±0.6	1.1±0.2	1.0±0.2	1.1±0.2	1.1±0.2	1.2±0.2	1.2±0.3	1.2±0.2
Current smoker (%)	13	15	14	13	14	12	9	8
Using medication for CVD or HTN (%)	41	60	43	68	54	71	39	82

[a] By Mono-S standard. Only measured in approximately 60% of the subjects.

Table 7. Baseline characteristics of the AGES-RS echocardiography subcohort (Paper I).

	Men (n=378)				Women (n=463)			
	NFG	**IFG**	**New-onset diabetes**	**Established diabetes**	**NFG**	**IFG**	**New-onset diabetes**	**Established diabetes**
Number (n)	251	60	15	52	349	62	12	40
Mean age (years)	76±6	76±5	75±6	76±5.2	76±6	77±7	77±4	77±5
SBP (mmHg)	141±20	142±20	147±15	147±21	142±20	142±21	145±18	144±21
DBP (mmHg)	76±10	78±9	82±9	74±8	73±9	72±9	76±7	71±8
BMI (kg/m^2)	26±4	28±4	29±4	28±4	27±5	29±5	29±6	30±6
Total cholesterol (mmol/l)	5.3±1.0	5.2±1.0	5.4±1.5	4.6±1.0	6.0±1.0	6.3±1.2	6.3±1.0	5.7±1.4
LDL (mmol/l)	3.4±0.9	3.2±0.9	3.4±1.4	2.7±1.0	3.8±1.0	4.1±1.1	3.8±0.8	3.4±1.2
HDL (mmol/l)	1.5±0.4	1.5±0.5	1.3±0.3	1.2±0.3	1.7±0.4	1.6±0.4	1.7±0.4	1.5±0.4
TG (mmol/l)	1.1±0.5	1.2±0.5	1.6±0.9	1.5±0.9	1.1±0.5	1.4±0.6	1.7±0.7	1.7±0.9
s-creatinine (mg/dl)	100±21	104±23	105±24	105±25	84±23	87±21	88±14	93±26
s-FG (mmol/l)	5.5±0.3	6.4±0.2	7.5±0.4	8.1±2.0	5.3±0.4	6.4±0.2	8.8±3.2	7.2±1.9
HbA1c (%)[a]	4.6±0.3	4.8±0.3	5.1±0.3	5.6±0.8	4.6±0.3	4.8±0.3	5.6±1.9	5.3±0.7
Current smoker (%)	12	10	0	16	10	7	17	5
Using medication for CVD or HTN (%)	62	77	87	87	63	73	92	85

[a] Measured by DCCT standard, converted to MonoS standard by using the following equation: ([HbA1c by DCCT] *1.03)-1.18, provided by EQUALIS (External Quality Assurance in Laboratory Medicine in Sweden).

Table 8. Baseline characteristics of the whole MPP-RES cohort (Papers III-IV).

	Middle-aged (57-69 years)				Elderly (70-86 years)			
MEN	**NFG**	**IFG**	**New-onset diabetes**	**Established diabetes**	**NFG**	**IFG**	**New-onset diabetes**	**Established diabetes**
Number (n)	4977	1299	158	721	3002	777	85	527
Mean age (years)	64±3	64±4	63±3	65±3	75±3	75±3	75±3	75±3
SBP (mmHg)	143±19	150±19	157±20	148±19	145±19	152±20	152±23	148±20
DBP (mmHg)	84±11	88±10	90±10	84±10	83±11	85±10	86±11	81±11
BMI (kg/m^2)	27±4	28±4	30±5	30±5	26±3	28±4	29±3	28±4
Waist circumference (cm)	97±10	102±11	108±12	105±12	97±10	102±10	104±9	103±11
Total cholesterol (mmol/l)	5.5±1.0	5.6±1.1	5.7±1.1	4.8±1.1	5.4±1.0	5.3±1.0	5.3±1.0	4.7±1.0
LDL (mmol/l)	3.7±0.9	3.7±1.0	3.7±1.0	3.0±0.9	3.5±0.9	3.4±0.9	3.3±0.9	2.8±0.9
HDL (mmol/l)	1.3±0.4	1.2±0.3	1.1±0.3	1.1±0.3	1.4±0.4	1.3±0.4	1.3±0.3	1.2±0.3
TG (mmol/l)	1.2±0.8	1.5±1.1	2.1±1.6	1.7±1.3	1.1±0.6	1.4±0.8	1.7±0.9	1.5±0.9
FPG (mmol/l)	5.4±0.4	6.5±0.5	9.1±2.7	8.6±2.7	5.3±0.4	6.5±0.5	8.9±2.3	8.4±2.5
Current smoker (%)	21	21	25	17	14	13	15	10
Physically inactive (%)	9	11	22	17	9	12	16	19
Using medication for CVD or HTN (%)	35	44	45	75	53	62	65	84

Table 8, continued.

	Middle-aged (57-69 years)				Elderly (70-80 years)			
WOMEN	**NFG**	**IFG**	**New-onset diabetes**	**Established diabetes**	**NFG**	**IFG**	**New-onset diabetes**	**Established diabetes**
Number (n)	2979	331	28	202	2517	333	45	245
Mean age (years)	66±4	66±3	67±2	67±3	73±3	74±3	74±3	74±3
SBP (mmHg)	140±20	146±20	147±23	145±21	146±21	151±22	157±24	146±20
DBP (mmHg)	82±10	85±11	86±9	82±10	82±10	85±11	87±11	81±10
BMI (kg/m^2)	26±4	29±5	31±6	30±6	37±4	29±5	30±5	30±5
Waist circumference (cm)	86±11	93±12	100±13	97±14	86±11	93±12	97±11	96±13
Total cholesterol (mmol/l)	6.0±1.0	6.0±1.0	6.2±1.4	5.3±1.1	6.0±1.1	5.9±1.1	6.2±1.2	5.1±1.0
LDL (mmol/l)	3.9±1.0	3.9±0.9	4.1±1.3	3.2±0.9	3.8±1.0	3.7±1.0	4.0±1.0	3.0±0.9
HDL (mmol/l)	1.6±0.4	1.4±0.4	1.2±0.3	1.4±0.4	1.7±0.4	1.5±0.5	1.3±0.4	1.4±0.4
Triglycerides (mmol/l)	1.1±0.6	1.5±0.7	2.1±0.9	1.6±1.6	1.2±0.5	1.4±0.7	1.8±1.0	1.6±0.9
FPG (mmol/l)	5.2±0.4	6.5±0.4	9.6±3.5	8.5±3.0	5.2±0.4	6.5±0.5	8.7±2.2	8.4±2.4
Current smoker (%)	19	21	21	14	12	15	11	11
Physically inactive (%)	8	11	21	19	12	20	27	20
Using medication for CVD or HTN (%)	32	51	69	48	48	59	49	82

Table 9. Results of the multivariable regression analysis of data from the AGES-RS echocardiography subcohort. Raw (unadjusted) mean values ± 1 SD are given for LV function/structure parameters for the various glucometabolic groups.

	s-FG ≤5.0	5.1-5.5	5.6-6.0	IFG	New-onset diabetes	Established diabetes	p-trend A[a]	p-trend F[b]
Em (cm/s)	7.8±1.5	8.1±1.7	8.0±1.7	7.9±1.5	7.8±1.7	8.1±2.9	0.8	0.5
Am (cm/s)	10.8±1.8	10.9±2.0	11.0±2.0	11.2±2.1	10.7±1.8	11.0±2.3	0.6	0.4
Em/Am	0.74±0.18	0.76±0.22	0.74±0.20	0.71±0.14	0.71±0.16	0.74±0.21	0.2	0.3
Transmitral E (cm/s)	72.3±17.9	70.8±16.9	72.9±17.6	75.8±20.3	72.3±22.1	75.9±18.7	0.2	0.3
Transmitral A (cm/s)	82.3±21.8	80.8±20.0	82.0±19.1	86.9±24.4	81.4±15.0	89.1±26.0	0.1	0.4
E/A	0.90±0.21	0.90±0.25	0.91±0.26	0.88±0.21	0.85±0.27	0.90±0.35	0.3	0.6
E /Em	9.7±2.8	9.2±2.9	9.6±3.0	9.8±3.2	9.5±2.8	10.0±3.0	0.4	0.97
LA diameter (mm/m^2)	2.14±0.36	2.09±0.28	2.11±0.34	2.07±0.29	2.09±0.26	2.10±0.29	0.3	0.1
LVMI (g/m^2)	89±29	85±21	90±25	90±23	95±21	91±19	0.06	0.4

[a]Age-adjusted, [b] fully adjusted. p-values from the trend tests are displayed (far right), performed on the first five groups only (excluding subjects with established diabetes).

No such trends or correlations were seen in women. The correlation between HbA1c and LVMI remained significant only for women (age-adjusted *F*-statistic 8.5; p=0.004; fully adjusted *F*-statistic 9.2; p=0.003).

The MPP-RES cohort

Table 11 gives the raw mean values (±1 SD) and adjusted intergroup trends for LV function/structure variables for the glucometabolic groups. Subjects with prevalent CHD, HF and significant valvular disease (n=190) were excluded from all calculations. There was a significant trend towards increasing LV diastolic dysfunction with increasing glucometabolic impairment, as evidenced by increased Am (fully adjusted), transmitral A (fully adjusted) and reduced Em/Am (age-adjusted). A significant trend towards increasing transmitral E with greater glucometabolic impairment was also seen (fully adjusted). In the gender-specific analysis the trends for the transmitral variables were stronger in men, while the trends in TDI variables were stronger in women.

Table 10. Associations between s-FG, HbA1c and HOMA and LV function/structure variables in the AGES-RS echocardiography subcohort, expressed in terms of the *F*-statistic and p-values.

		s-FG		**HbA1c**		**HOMA**	
		F-statistic	p	*F*-statistic	p	*F*-statistic	p
Em (cm/s)	A[a]	-0.6	0.4	-2.9	0.09	-2.1	0.2
	F[b]	-0.02	0.9	-0.4	0.5	-0.5	0.5
Am (cm/s)	A	+0.07	0.8	-5.9	**0.02**	-0.01	0.9
	F	+0.7	0.4	-0.5	0.5	-0.2	0.7
Em/Am	A	-2.2	0.1	-0.2	0.7	-2.8	0.1
	F	-1.6	0.2	-0.3	0.6	-2.7	0.1
Transmitral E (cm/s)	A	+0.9	0.4	+0.002	1.0	+7.7	**0.006**
	F	+0.2	0.7	+1.5	0.2	+1.3	0.2
Transmitral A (cm/s)	A	+0.2	0.6	+0.005	0.9	+10.2	**0.001**
	F	+0.01	0.9	+0.1	0.7	+3.1	0.08
E/A	A	-0.1	0.7	-0.4	0.5	-1.1	0.3
	F	-0.1	0.7	-2.0	0.2	-2.5	0.1
E /Em	A	+0.6	0.4	+0.8	0.4	+10.9	**0.001**
	F	+0.02	0.9	+0.06	0.8	+3.7	0.06
LAD (mm/m^2)	A	+0.06	0.8	+3.7	0.06	+0.2	0.6
	F	+0.1	0.7	+0.3	0.6	+0.9	0.3
LVMI (g/m^2)	A	+3.9	**0.049**	+8.8	**0.003**	+3.0	0.08
	F	+1.6	0.2	+10.7	**0.001**	+2.0	0.2

[a]Age-adjusted, [b] fully adjusted. Plus (+) and minus (-) signs denote whether the associations are positive or negative.

Table 11. Results of the multivariable regression analysis of the data from the MPP-RES echocardiography subcohort. The values are given as raw (unadjusted) means ± 1 SD.

	FPG ≤5.0	**5.1-5.5**	**5.6-6.0**	**IFG**	**New-onset diabetes**	**Established diabetes**	**p-trend**	
							A[a]	**F**[b]
Em (cm/s)	7.6±2.5	7.9±2.8	8.4±2.8	8.2±2.8	8.2±2.7	7.4±2.6	0.8	0.1
Am (cm/s)	10.8±2.7	11.6±2.8^{*A,*F}	12.4±2.7$^{‡A,\ ‡F}$	12.3±2.8^{‡A,§F}	12.2±3.0^{‡A,†F}	11.3±2.8^{*F}	0.0002	0.0004
Em/Am	0.72±0.24	0.68±0.21^{*A}	0.68±0.18^{†A,*F}	0.68±0.19^{‡A,†F}	0.68±0.18^{*A}	0.65±0.18^{‡A,†F}	0.006	0.3
Transmitral E (cm/s)	72.2±16.0	68.2±14.7^{*A}	70.1±14.9	70.2±17.4^{†A}	74.5±16.9	73.6±17.9^{†A,*F}	0.04	0.02
Transmitral A (cm/s)	78.7±16.8	76.3±16.5	76.0±15.2	78.5±17.3	81.0±17.6^{*A}	85.1±18.5^{§A,§F}	0.0002	0.03
E/A	0.95±0.26	0.92±0.24	0.95±0.27	0.92±0.24	0.95±0.26	0.89±0.24	0.2	0.8
E/Em	10.6±4.1	9.5±3.3	9.1±3.5	9.4±4.0	10.1±3.9	11.0±4.2	0.2	0.9
LAD (mm/m^2)	20.6±2.7	20.8±2.7	20.5±2.7	20.7±2.7	20.6±2.6	21.1±2.6	0.5	0.6
LVMI (g/m^2)	81±20	89±24^{§A,†F}	87±19^{‡A,*F}	88±21^{§A,*F}	89±21^{‡A}	91±23^{§A}	0.001	0.7

[a]Age-adjusted, [b] fully adjusted. Significant intergroup differences were calculated using the first group (FPG ≤5.0 mmol/l) as reference group. p-values from the trend tests are displayed (far right), performed on the first five groups only (excluding subjects with established diabetes).
$^{*}p<0.05$; $^{†}p<0.01$; $^{‡}p<0.001$; $^{§}p<0.0001$.

Associations between FPG and HbA1c and LV function/structure variables are given in Table 12. Positive associations were found between FPG and Am, transmitral A and transmitral E in the age-adjusted models, but these were weaker or disappeared in the fully adjusted models. Weaker associations were generally seen with HbA1c. No gender differences were observed. There was a significant trend toward increasing LMVI with increasing glucometabolic impairment (age-adjusted). FPG was positively correlated with LVMI (age-adjusted). In the gender specific analysis the intergroup trend was seen only in women (fully adjusted).

Table 12. Associations between FPG and HbA1c and LV function/structure variables in the MPP-RES echocardiography subcohort expressed in terms of the *F*-statistic and p-values.

		FPG		**HbA1c**	
		F-statistic	p	*F*-statistic	p
Em (cm/s)	A[a]	+0.02	0.9	-0.001	1.0
	F[b]	+1.1	0.3	-0.4	0.5
Am (cm/s)	A	+7.0	**0.008**	+4.1	**0.04**
	F	+3.6	0.06	+2.1	0.1
Em/Am	A	-3.6	0.06	-2.5	0.1
	F	-0.06	0.8	-0.3	0.6
Transmitral E (cm/s)	A	+4.8	**0.03**	+3.64	0.06
	F	+5.9	**0.02**	+3.0	0.08
Transmitral A (cm/s)	A	+13.8	**0.0002**	+12.7	**0.0004**
	F	+3.1	0.08	+4.5	**0.03**
E/A	A	-1.3	0.3	-1.7	0.2
	F	-0.2	0.7	-0.09	0.8
E /Em	A	+2.1	0.2	+3.5	0.06
	F	+0.4	0.5	+1.8	0.2
LAD (mm/m^2)	A	-0.1	0.7	+0.07	0.8
	F	-0.5	0.5	+0.2	0.7
LVMI (g/m^2)	A	+4.5	**0.03**	+0.06	0.8
	F	+0.08	0.8	+0.9	0.4

[a]Age-adjusted, [b] fully adjusted. Plus (+) and minus (-) signs denote whether the associations are positive or negative.

In the MPP-RES post hoc age-stratified analysis the significant trends of increasing LVDD with increasing glucometabolic impairment observed for the whole cohort remained significant for Am (full adjustment), Em/Am (age-adjustment) and transmitral A (age-adjusted) in the middle-aged group but not in the elderly group (Table 13). Likewise, the age-adjusted positive correlation between FPG and Am, transmitral A and transmitral E remained significant only in the middle-aged group. There were differences in LVMI between glucometabolic groups in both age groups, but the intergroup trend towards increasing LVMI with increasing glucometabolic impairment (age- and fully adjusted) and the association between FPG and LVMI (age-adjusted) remained significant only in the elderly group. The association between HbA1c and transmitral A was stronger in the elderly group (fully adjusted). In the gender-stratified analyses the observed associations remained significant most often among middle-aged men who comprised the largest group (middle-aged men n=772, middle-aged women n=208, elderly men n=279, elderly women n=260). p-values were generally of borderline significance (0.02-0.04).

Paper II

Nt-proBNP was on average higher in women and in subjects with prevalent CHD and HF; and lower in overweight and obese subjects (adjusted for age and cystatin C). Results from the linear regression analysis are given in Table 14. When all the subjects in the MPP-RES echocardiography subcohort were included, a positive correlation was found between Nt-proBNP and FPG among middle-aged and elderly men, although this was statistically significant only in the unadjusted and fully adjusted models for middle-aged men. Among middle-aged women, a statistically significant inverse correlation was observed after adjustment for age and cystatin C. Significance was lost with further adjustments but the *t*-statistic remained negative. A negative correlation was also observed among the elderly women, although it was not significant with any kind of adjustment. A similar pattern was observed after excluding subjects with established diabetes, prevalent CHD, HF or significant valvular disease (n=626) (Table 14). Variables remaining significant in the fully adjusted models are given in Table 15.

Table 13. Results from the post hoc multivariable regression analysis of the data from the MPP-RES echocardiography subcohort, for middle-aged (57-69 years) and elderly (70-80 years) individuals. The values are given as raw (unadjusted) means ± 1 SD.

	FPG ≤5.0	**5.1-5.5**	**5.6-6.0**	**IFG**	**New-onset diabetes**	**Established diabetes**	**p-trend**	
							A[a]	**F**[b]
Middle-aged (n)	64	137	159	315	109	196		
Em (cm/s)	8.4±2.5	8.6±2.8	8.9±2.6	8.8±2.6	8.8±2.7	8.1±2.6	0.9	0.1
Am (cm/s)	11.7±2.8	12.0±2.6	12.8±2.6^{*A,*F}	12.8±2.6$^{*A,\dagger F}$	12.8±2.8$^{\dagger A,*F}$	12.0±2.9	0.001	0.002
Em/Am	0.74±0.23	0.72±0.19	0.70±0.18^{*A,*F}	0.69±0.19$^{\dagger A,*F}$	0.69±0.17^{*A}	0.69±0.19^{*A}	0.009	0.4
Transmitral E (cm/s)	70.7±14.0	68.3±13.1	69.3±14.1	69.2±16.2	74.6±16.5	74.3±17.5	0.04	0.02
Transmitral A (cm/s)	74.8±16.0	72.1±15.0	74.1±14.2	76.2±16.3	77.3±14.9	82.2±17.1$^{\ddagger A,\ddagger F}$	0.001	0.1
E/A	0.98±0.24	0.98±0.24	0.97±0.26	0.93±0.23	0.99±0.27	0.93±0.26	0.2	0.7
E/Em	9.2±3.6	8.6±2.9	8.4±2.8	9.4±4.0	10.0±3.7	11.7±4.1^{*A}	0.2	0.5
LAD (mm/m^2)	20.1±2.2	20.6±2.7	20.3±2.6	20.4±2.5	20.3±2.5	20.5±2.3	1.0	0.4
LVMI (g/m^2)	81.1±19.1	87.5±20.5^{*A,*F}	85.6±18.5^{*A}	85.6±19.8^{*A}	87.5±21.1^{*A}	91.6±24.4$^{\ddagger A}$	0.2	0.2

[a]Age-adjusted, [b] fully adjusted.

Table 13, continued

	FPG ≤5.0	5.1-5.5	5.6-6.0	IFG	New-onset diabetes	Established diabetes	p-trend A[a]	p-trend F[b]
Elderly (n)	70	87	54	121	43	164		
Em (cm/s)	6.8±2.1	6.7±2.3^{*A,*F}	7.1±2.9^{*A,*F}	6.7±2.5$^{*A,\dagger F}$	6.6±2.1	6.4±2.2	0.8	0.6
Am (cm/s)	10.0±2.3	10.9±3.0^{*A,*F}	11.1±2.9^{*A,*F}	11.0±2.9$^{*A,\dagger F}$	10.5±2.8	10.5±2.5	0.2	0.08
Em/Am	0.69±0.24	0.62±0.21^{*A}	0.64±0.15	0.62±0.21^{*A}	0.65±0.18	0.61±0.16$^{\dagger A,\dagger F}$	0.2	0.6
Transmitral E (cm/s)	73.5±17.7	68.2±16.9	72.4±17.2	72.9±20.1	74.4±18.0	72.9±18.5	0.3	0.3
Transmitral A (cm/s)	82.3±16.8	83.3±16.8	81.8±16.6	84.7±18.3	90.6±20.5^{*A}	88.5±19.6^{*A,*F}	0.06	0.2
E/A	0.92±0.29	0.82±0.21^{*A,*F}	0.91±0.28	0.88±0.27	0.84±0.22	0.83±0.21^{*A}	0.8	1.0
E/Em	11.7±4.1	11.1±3.4	11.2±4.4	12.1±5.3	11.9±3.3	12.3±4.3^{*A,*F}	0.3	0.5
LAD (mm/m2)	21.0±3.0	21.3±2.7	21.1±2.7	21.8±2.7	21.3±2.7	21.8±2.7	0.3	1.0
LVMI (g/m^2)	80.9±20.1	92.1±28.5$^{\dagger A,\dagger F}$	90.1±19.0$^{\dagger A}$	96.1±22.0$^{*A,\ddagger F}$	91.5±19.3$^{\dagger A}$	89.6±21.3$^{\dagger A}$	0.0004	0.03

[a]Age-adjusted, [b] fully adjusted. Significant intergroup differences were calculated using the first group (FPG ≤5.0 mmol/l) as reference group). p-values from the trend tests are displayed (far right), performed on the first five groups only (excluding subjects with established diabetes).
$^{*}p<0.05$; $^{\dagger}p<0.01$; $^{\ddagger}p<0.001$.

Table14. Correlation between Nt-proBNP and FPG in the MPP-RES echocardiography subcohort.

All subjects included	***t*-statistic**			
(n=1775)	**Middle-aged**		**Elderly**	
Variables adjusted for:	Men (n=866)	Women (n=229)	Men (n=392)	Women (n=288)
Unadjusted	2.07*	-1.80	1.69	-0.51
Age	1.86	-2.76†	1.69	-.070
Age, cystatin C	1.79	-2.88†	1.67	-1.15
Age, cystatin C, BMI	1.74	-1.80	1.69	-0.65
Age, cystatin C, CVD risk factors	1.84	-1.30	1.49	0.00
Age, cystatin C, CVD risk factors, echocardiography variables and AF	2.03*	-1.47	1.25	-0.51
Subjects with established diabetes, CHD, HF or significant valvular disease excluded				
(n=1149)	**Middle-aged**		**Elderly**	
	Men (n=605)	Women (n=174)	Men (n=182)	Women (n=187)
Unadjusted	0.79	-2.24*	2.37*	-1.74
Age	0.80	-2.94†	2.20*	-1.74
Age, cystatin C	1.02	-2.82†	2.23*	-1.72
Age, cystatin C, BMI	0.91	-1.47	2.66†	-0.92
Age, cystatin C, CVD risk factors	1.26	-1.19	2.95†	0.45
Age, cystatin C, CVD risk factors, echocardiography variables and AF	2.97†	-1.03	2.57*	-0.30

*p<0.05; †p<0.01.

Substituting FPG for HbA1c in the regression analysis resulted in a positive *t*-statistic among men and a negative *t*-statistic among women for all kinds of adjustment. However, the correlations were not significant for either men or women, in the whole echocardiography subcohort or after excluding subjects with established diabetes, CHD, HF or valvular disease.

Table 15. Covariates remaining significantly associated with Nt-proBNP in the fully adjusted regression model. Subjects with established diabetes, CHD, HF or significant valvular disease excluded.

	***t*-statistic**			
	Middle-aged		**Elderly**	
	Men	Women	Men	Women
Age	3.73‡	6.35§	3.48‡	-
FPG	2.97†	-	2.57*	-
Cystatin C	-	5.34§	3.17†	3.69‡
BMI	-2.62†	-2.71†	-	-
LVMI	5.72§	-	2.01*	-
Em	2.60*	-	-	-
Am	-5.30§	-	-	-2.98†
Sm	-	-	-2.35*	-
LVEF	-	-	2.04*	-
Using medication for CVD or hypertension	4.06§	-	-	-
s-TG	-	-	-2.27*	-2.53*
Mean pulse rate	-2.05*	-	-	-2.91†

When no *t*-statistic is given (-), the relevant covariate did not reach statistical significance and was thus excluded from the regression analysis. *p<0.05; †p<0.01; ‡p<0.001; §p<0.0001.

Raw (unadjusted) mean Nt-proBNP values in the glucometabolic groups in both age groups are shown in Figure 5. After adjusting for all covariates, middle-aged men with new-onset and established diabetes had significantly higher Nt-proBNP levels than the reference group (FPG≤5.0 mmol/l) (9.53 pmol/l (p=0.002) and 8.23 pmol/l (p=0.02) vs. 5.71 pmol/l). No significant differences were observed between the glucometabolic groups among elderly men or among women of either age group. Excluding subjects with prevalent CHD, HF or significant valvular disease did not change the results, with the exception that women with established diabetes had a lower mean Nt-proBNP level than the reference group (7.46 pmol/l vs. 12.81 pmol/l; p=0.006).

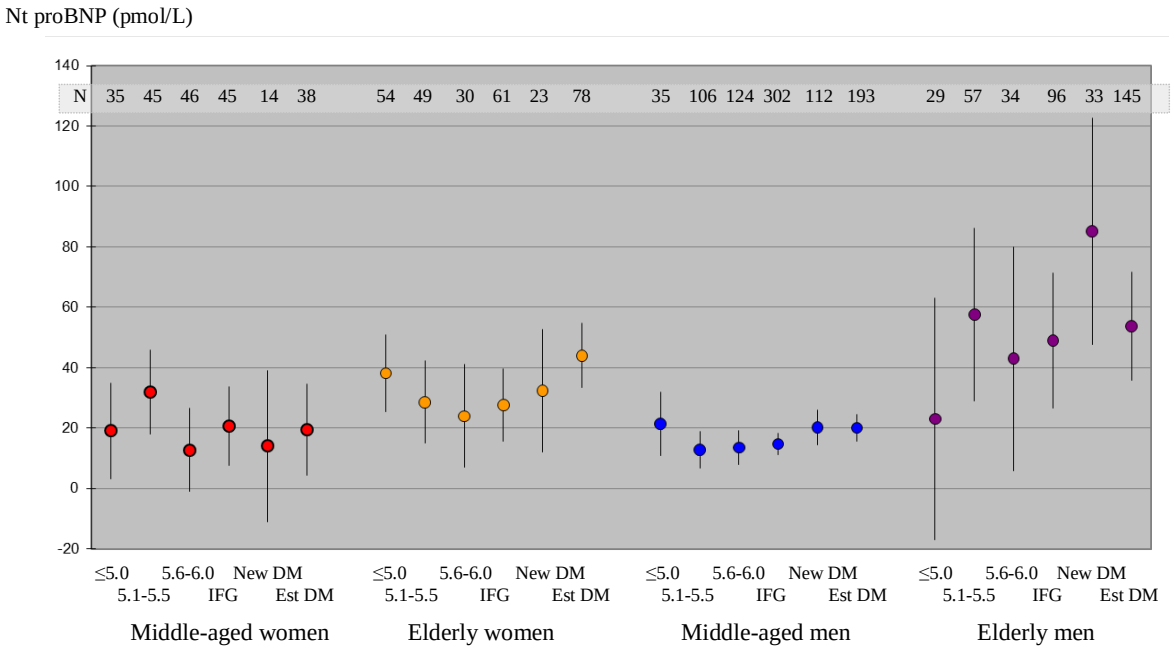

Figure 5. Raw (unadjusted) mean Nt-proBNP levels (pmol/l) as a function of glucometabolic impairment. Vertical bars represent 95% CIs. New DM = new-onset type 2 diabetes; IFG = impaired fasting glucose; Est DM = established diabetes.

In an attempt to explain the observed gender difference in the association between Nt-proBNP and FPG, a post hoc analysis was performed to assess the degree of explanation (R^2) for the variance in Nt-proBNP. When all the subjects in the MPP-RES echocardiography subcohort were included, 52.3% of the variance for men and 40.2% for women could be explained by the variables in the model. The strongest correlation was found for age for both sexes (R^2 for men 25.7% and for women 14.3%). Cystatin C explained 5.4% and 11.3% of the variance for men and women, respectively. Together, LV diastolic function variables explained 14.4% of the variance for men and 9.7% for women. R^2 for LVEF was 1.4% among men and 0.5% among women. Similarly, R^2 for LVMI was higher among men (2.7%) than women (0.3%). R^2 for each of the remaining variables in the model was <1.0%. The gender difference regarding echocardiographic variables was similar after excluding subjects with established diabetes, prevalent CHD, HF or significant valvular disease, and also when the middle-aged and elderly age groups were analysed separately.

Paper III

Risk factor burden

The proportions (%) of subjects within each glucometabolic group and age group in the whole MPP-RES cohort (n=18,238) having at least three of the five risk factors chosen (uncontrolled hypertension, dyslipidaemia, central obesity, current smoking or lack of physical activity) can be seen in Figure 6. For the whole cohort and each

glucometabolic group, women in general exhibited more risk factors than men, also after adjustment for age. There was a highly significant trend among both men and women for increasing risk factor burden with increasing glucometabolic impairment in both age groups (p-trend <0.0001). The ratio between the β-coefficients from the trend tests (reflecting the slope of the curve) for middle-aged vs. elderly men was 1.21 (p-interaction=0.12) and for women 0.97 (p-interaction=0.78).

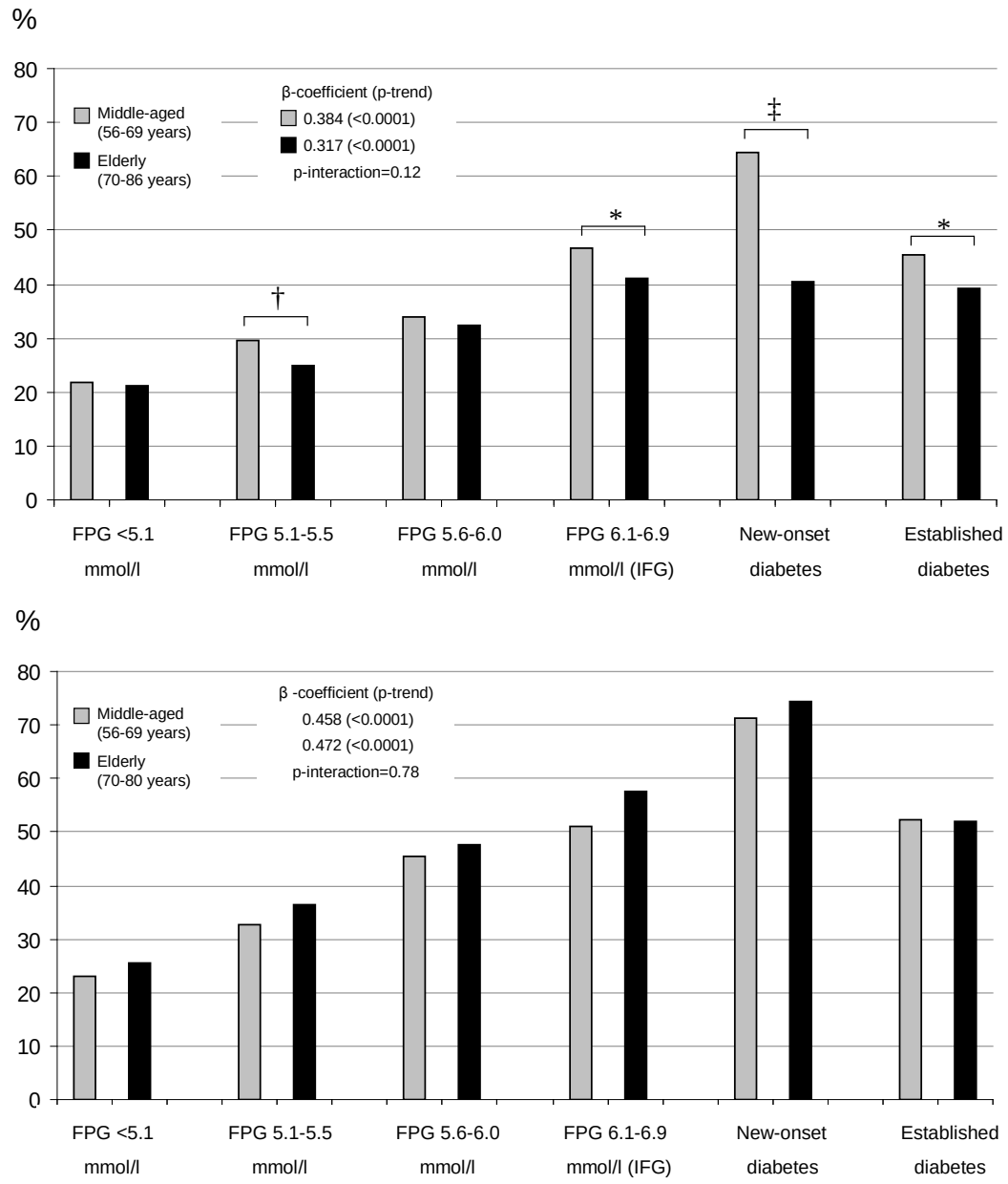

Figure 6. Proportion of men (above) and women (below) with three or more risk factors – uncontrolled hypertension, dyslipidaemia, central obesity, current smoker or being physically inactive – for each of the six glucometabolic groups. β-coefficients and p-values for the intergroup trends are also shown. Brackets denote significant differences between age groups within each glucometabolic group. $^{*}p<0.05$; $^{\dagger}p<0.01$; $^{\ddagger}p<0.001$.

Although the difference in trends between age groups was not significant among men, elderly men with FPG 5.1-5.5 mmol/l (OR 0.78 [95% CI 0.67-0.92], p=0.002), IFG (0.80 [0.67-0.96], p=0.02), new-onset diabetes (0.37 [0.22-0.65], p=0.0004) and established diabetes (0.78 [0.62-0.98], p=0.03) had significantly fewer risk factors than the middle-aged men in the corresponding groups. No such differences were seen between age groups among the women. The ratio between β-coefficients for middle-aged women vs. middle-aged men was 1.19 (p-interaction=0.1) while for elderly women vs. elderly men it was 1.49 (p-interaction=0.002).

Self-rated health

The proportions (%) of subjects within each glucometabolic group and age group reporting low SRH are presented in Figure 7. On average, women and elderly subjects of both sexes more often reported low SRH. Middle-aged men with NFG (the first three groups) and IFG (fourth group) reported significantly better SRH than elderly men in these groups, while there was no difference in SRH between middle-aged and elderly men with new-onset and established diabetes. Consequently, the trend with increasing glucometabolic impairment was stronger in the middle-aged men (β-coefficient=0.21, p-trend <0.0001) than in the elderly men (β-coefficient=0.11, p-trend <0.0001), generating a ratio of 1.89 (p-interaction=0.005). For women, the β-coefficient for middle-aged women was 0.17 (p-trend <0.0001) compared to 0.14 (p-trend <0.0001) for elderly women, the ratio being 1.22 (p-interaction=0.97). The differences in β-coefficients between men and women in each age group were not statistically significant (middle-aged: β-coefficient ratio 0.82, p-interaction=0.3; elderly: β-coefficient ratio 1.26, p-interaction=0.5).

Paper IV

In the whole MPP-RES cohort (n=18,238) the mean follow-up period was 3.9±1.3 years for men and 4.5 ±1.3 years for women, during which 972 and 324 CVD events occurred, respectively. Eighteen men and seven women were lost to follow-up, the most common reason being emigration from the country.

HRs for CVD events for subjects divided into glucometabolic groups, using the first group (FPG ≤5.0 mmol/l) as reference, together with p-values for intergroup trends from the first to the fifth group are shown in Figures 8 (men) and 9 (women) for the two age groups. In the unadjusted analysis a significant trend was seen towards higher HRs with increasing glucometabolic impairment for the middle-aged men (β-coefficient 0.20, p-trend=0.0001) and middle-aged women (β-coefficient 0.30, p-trend=0.002). In the fully adjusted analysis the trend was weaker for both sexes, remaining significant for middle-aged men only (0.14 for men, p-trend=0.01; 0.13 for women, p-trend=0.22). The β-coefficient for the trend was similar for both sexes, but among the women the confidence intervals overlapped 1.0 due to the smaller number of events, rendering the trend insignificant.

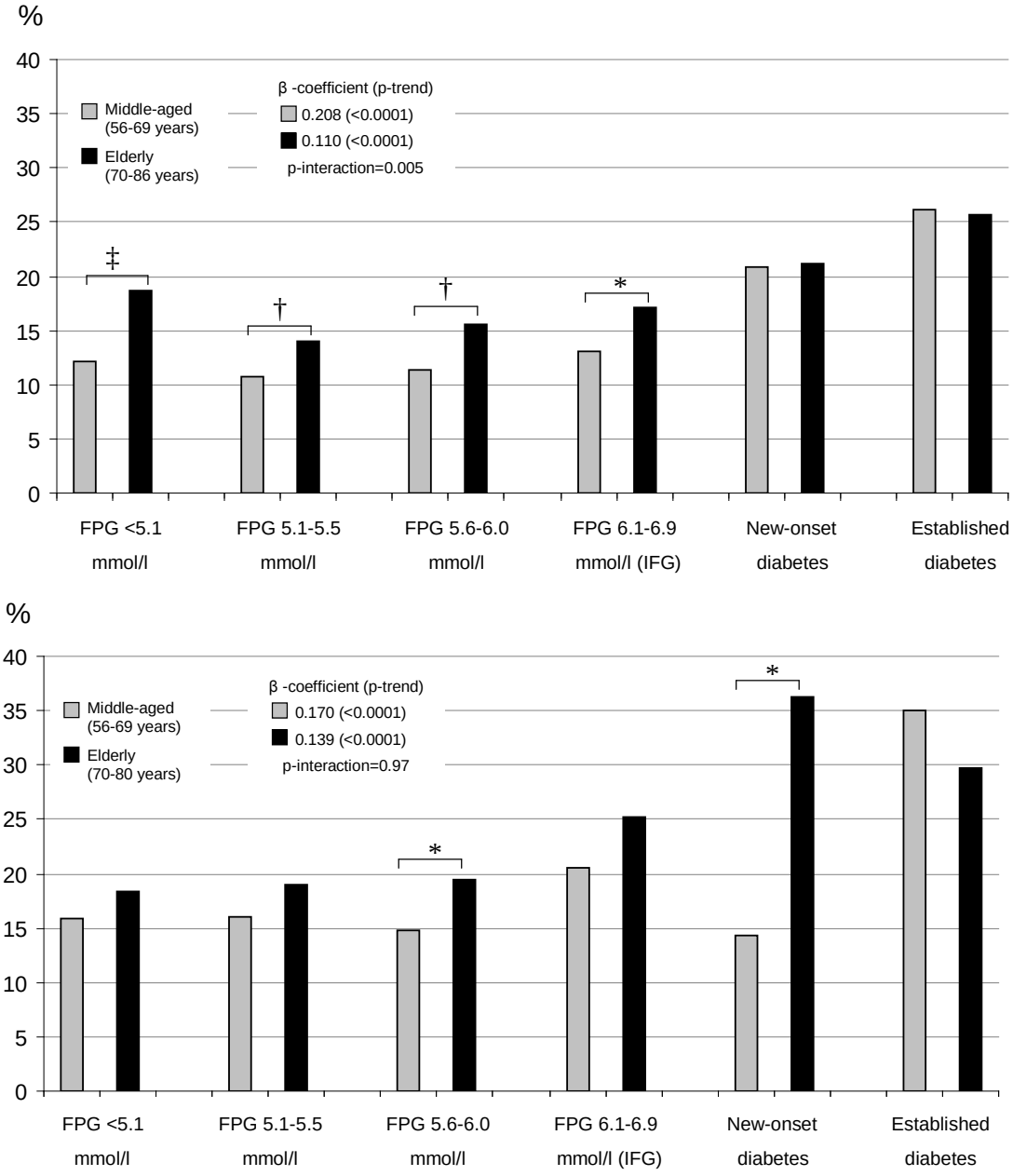

Figure 7. Proportion of men (above) and women (below) reporting low SRH within each of the six glucometabolic groups. β-coefficients and p-values for the intergroup trends are also shown. Brackets denote significant differences between age groups within each glucometabolic group.
.*p<0.05; †p<0.01; ‡p<0.001.

The HRs for the first four groups of elderly men (FPG ≤5.0-6.9 mmol/l) were close to 1.0, in both the unadjusted and the fully adjusted analyses (Figure 8). Consequently the trend from the first to the fifth groups among the elderly men was not significant (unadjusted or fully adjusted). For elderly women, the HRs for the first three groups (FPG ≤5.0-6.0 mmol/l) were close to 1.0 (Figure 9), while the HR for the fourth group (FPG 6.1-6.9 mmol/l) was 1.50 (0.97-2.32) in the unadjusted analysis. Further adjustments led to a weakening of the HR (1.24 (0.78-1.96)). The trend from the first

to the fifth groups among the elderly women was significant in the unadjusted model (p-trend=0.01) but significance was lost in the fully adjusted model (p-trend=0.27). The β-coefficients for the interaction between the trend from the first to the fifth groups and the two age groups for men were -0.17 (p-interaction=0.01, unadjusted) and -0.15 (p-interaction=0.03, fully adjusted), and for women -0.13 (p-interaction=0.27) in both models. As for the trend tests, however, due to fewer events among the women the confidence intervals were wide, explaining the lack of significance for women despite similar β-coefficients for the sexes. The three-way interaction analysis including sex thus showed no significant differences.

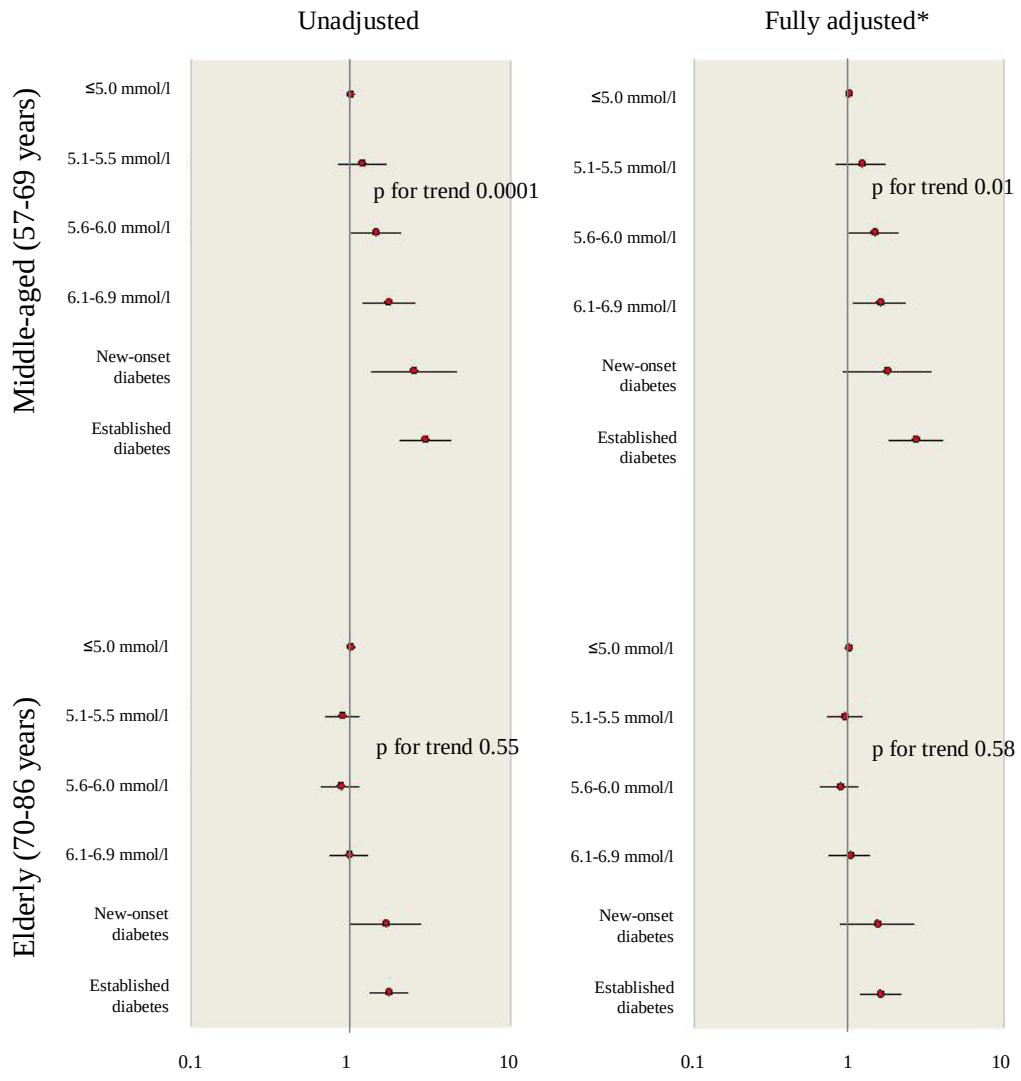

Figure 8. HRs and 95% CI bars for CVD events between the glucometabolic groups for men, using the first group (FPG ≤5.0 mmol/l) as a reference group. Subjects were divided into middle-aged (above) and elderly (below).
*Adjusted for: waist circumference, DBP, TG, HDL, current smoking, physical inactivity, prevalent CVD or heart failure and medication for CVD or hypertension.

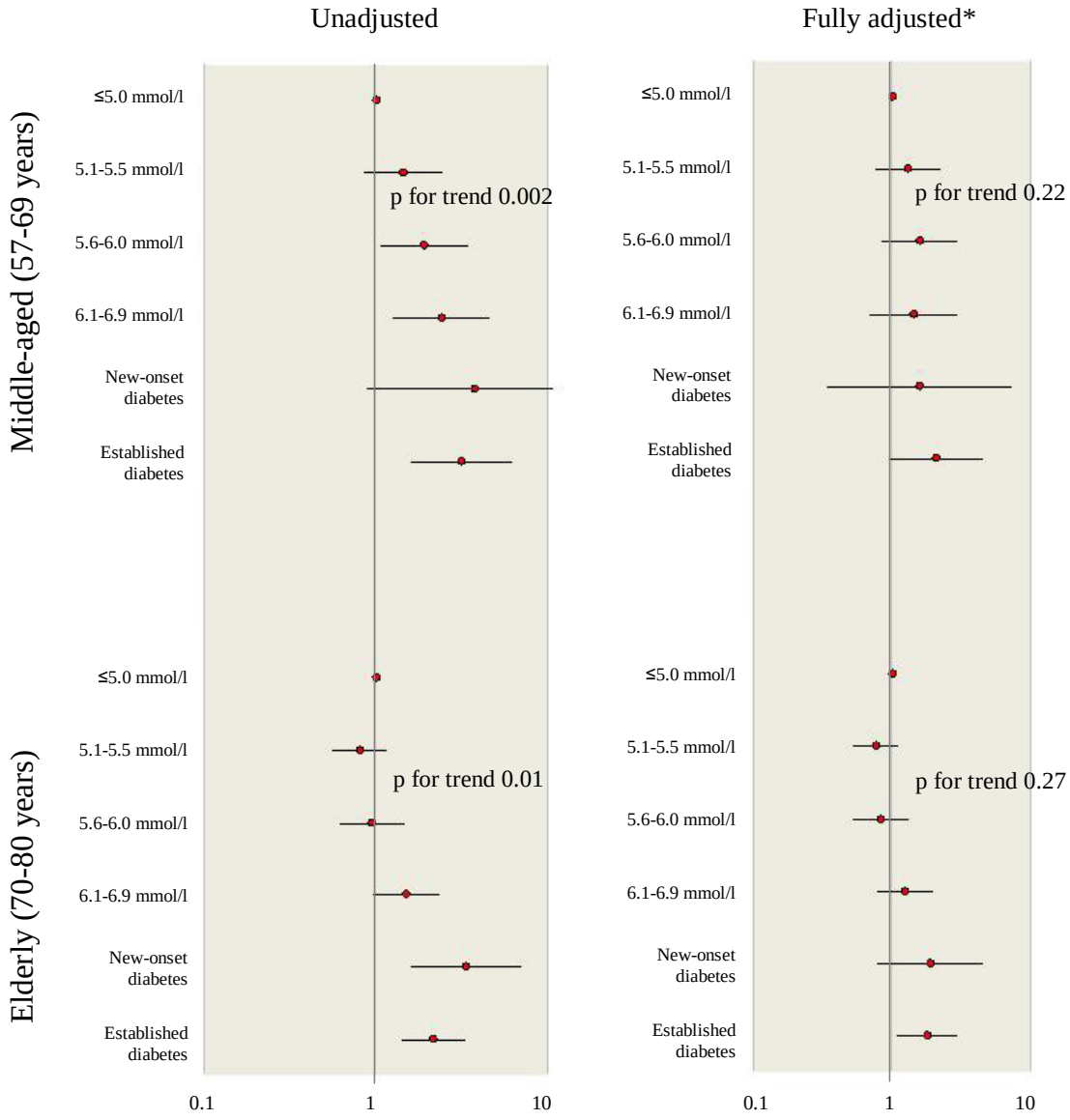

Figure 9. HRs and 95% CI bars for CVD events between the glucometabolic groups for women, using the first group (FPG ≤5.0 mmol/l) as a reference group. Subjects were divided into middle-aged (above) and elderly (below).
*Adjusted for: waist circumference, DBP, TG, HDL, current smoking, physical inactivity, prevalent CVD or heart failure and medication for CVD or hypertension.

Table 16 gives the numerical values for HRs and 95% CIs for those with impaired glucose metabolism (IFG by WHO criteria, new-onset diabetes and established diabetes). In the unadjusted models middle-aged and elderly women with new-onset diabetes in general had higher HRs for CVD events than men in the corresponding age groups, although the difference did not reach statistical significance (p-difference=0.06 for combined age groups). The difference was reduced in the fully adjusted model. For those with IFG and established diabetes, although the HRs

among women were slightly higher the difference between the sexes was less pronounced and non-significant (p-difference >0.10) in both the unadjusted and fully adjusted models.

Table 16. HRs and 95% CIs for middle-aged and elderly men and women with impaired glucose metabolism.

		MEN		**WOMEN**	
Unadjusted		HR	95% CI	HR	95% CI
Middle-aged	≤5.0 mmol/l	1.00 (ref)	-	1.00 (ref)	-
	IFG	1.72	1.19-2.50	2.40	1.25-4.60
	New-onset diabetes	2.48	1.33-4.63	3.75	0.88-15.92
	Established diabetes	2.92	2.00-4.25	3.15	1.60-6.22
Elderly	≤5.0 mmol/l	1.00 (ref)	-	1.00 (ref)	-
	IFG	0.97	0.73-1.29	1.50	0.97-2.32
	New-onset diabetes	1.63	0.97-2.73	3.35	1.60-6.99
	Established diabetes	1.72	1.30-2.27	2.16	1.40-3.32
*Fully adjusted**		HR	95% CI	HR	95% CI
Middle-aged	≤5.0 mmol/l	1.00 (ref)	-	1.00 (ref)	-
	IFG	1.57	1.07-2.32	1.42	0.69-2.92
	New-onset diabetes	1.75	0.90-3.41	1.56	0.34-7.20
	Established diabetes	2.66	1.78-3.98	2.06	0.95-4.44
Elderly	≤5.0 mmol/l	1.00 (ref)	-	1.00 (ref)	-
	IFG	1.00	0.74-1.35	1.24	0.78-1.96
	New-onset diabetes	1.51	0.88-2.61	1.88	0.78-4.54
	Established diabetes	1.59	1.17-.2.16	1.78	1.09-2.93

*Adjusted for waist circumference, DBP, TG, HDL, current smoking, physical inactivity, prevalent CVD or heart failure and medication for CVD or hypertension.

Additional unpublished results

In a further attempt to explain the results presented in Papers I and II unadjusted correlations between Nt-proBNP and selected echocardiographic variables reflecting LVDD and LVH were analysed, stratifying by age, sex and diagnosis of diabetes (new-onset or established diabetes) (Table 17). Nt-proBNP correlated well with LA area/m^2, Am, E/Em and LVMI for diabetic and non-diabetic men and women of both age groups. The weaker p-values for middle-aged diabetic women reflect the small size of the group, while the correlation coefficients were similar in all groups.

Table 17. Correlation (*r*) between Nt-proBNP and selected echocardiographic variables reflecting LVDD and LVH.

	MEN				**WOMEN**			
MIDDLE-AGED	**Non-diabetic (n=560)**		**Diabetic (n=303)**		**Non-diabetic (n=176)**		**Diabetic (n=52)**	
	r	p	*r*	p	*r*	p	*r*	p
LA-area/m^2	0.42	<0.0001	0.47	<0.0001	0.33	<0.0001	0.36	0.01
Em	-0.09	0.04	-0.2	0.001	-0.17	0.02	-0.07	0.6
Am	-0.33	<0.0001	-0.33	<0.0001	-0.37	<0.0001	-0.15	0.3
Em/Am	0.12	0.004	0.05	0.4	0.01	0.9	0.09	0.5
E/Em	0.22	<0.0001	0.34	<0.0001	0.29	<0.0001	0.27	0.06
LVMI	0.33	<0.0001	0.36	<0.0001	0.21	0.007	0.28	0.045
ELDERLY	**Non-diabetic (n=212)**		**Diabetic (n=178)**		**Non-diabetic (n=186)**		**Diabetic (n=97)**	
	r	p	*r*	p	*r*	p	*r*	p
LA-area/m^2	0.60	<0.0001	0.45	<0.0001	0.47	<0.0001	0.57	<0.0001
Em	-0.11	0.1	-0.11	0.2	-0.01	0.9	-0.28	0.009
Am	-0.37	<0.0001	-0.42	<0.0001	-0.28	<0.0001	-0.22	0.04
Em/Am	0.24	0.001	0.25	0.002	0.30	<0.0001	-0.11	0.3
E/Em	0.31	<0.0001	0.32	<0.0001	0.26	0.001	0.38	<0.0001
LVMI	0.35	<0.0001	0.38	<0.0001	0.11	0.1	0.32	0.002

DISCUSSION

Myocardial dysfunction

Possible associations between glucose metabolism and measures of myocardial dysfunction were examined using echocardiography (Paper I) and Nt-proBNP (Paper II). Only a few, and inconsistent, associations were observed between increasing glucometabolic impairment and measures of LVDD in the AGES-RS echocardiography subcohort and in the elderly group of the MPP-RES echocardiography subcohort, which in most cases lacked significance in the fully adjusted models. These observations contradict previous reports in younger subjects as well as the current findings in the middle-aged group in the MPP-RES cohort. A positive association between increasing glucometabolic impairment and increasing LVMI was seen in both cohorts, remaining significant for women only in the fully adjusted models.

A positive correlation was found between Nt-proBNP and FPG among middle-aged men, when analysing data from the MPP-RES echocardiography subcohort, including all and after excluding subjects with established diabetes, prevalent CHD, HF or significant valvular disease (Paper II). A positive correlation was also observed among elderly men albeit non-significant when all subjects were included. A non-significant negative correlation was, however, observed among women of both age groups. Middle-aged diabetic men also had higher mean Nt-proBNP values, while diabetic women had lower Nt-proBNP values than the reference groups (FPG $\leq$ 5.0 mmol/l). In a post hoc analysis it was found that only half of the variation in Nt-proBNP among men and less than half in the women could be explained by the covariates used in the model, i.e. age, cystatin C, CVD risk factors, medication and echocardiographic measures of LV structure and function. Nonetheless, in the complementary analyses it was found that the correlations between Nt-proBNP and echocardiographic measures of LVDD and LVH were strong among men and women of both age groups, regardless of whether they were diabetic or not.

Diastolic dysfunction and left ventricular hypertrophy

The majority of prior studies examining LV diastolic function in subjects with diabetes or the correlation of measures of LV diastolic function with continuous measures of glucose metabolism (i.e. FPG, 2-hour glucose, HbA1c or HOMA) have reported similar findings: namely a decrease in transmitral E and Em, a compensatory increase in transmitral A and Am, a subsequent decrease in E/A and Em/Am, an increase in E/Em and LA enlargement.[63, 64, 139, 140] Impaired relaxation of the LV myocardial wall is likely to be the explanation of these echocardiographic changes.[64] The majority of these studies have included middle-aged subjects with type 2 diabetes or young subjects with type 1 diabetes. There are few studies on the relationship between glucose metabolism and LV diastolic function in elderly subjects and these

have shown inconsistent results. The main studies found are summarized in Table 18. In a study by Andrén et al. of 70-year-old men in Uppsala, those with diabetes had a larger LA, but transmitral Doppler measurements of LVDD did not differ between healthy and diabetic subjects.[125] In the Cardiovascular Health Study by Lee et al. in elderly individuals aged 65-103 years, increased glucometabolic impairment was found to be positively correlated with transmitral E (men only) and A (both sexes).[127] A Portuguese group found that impaired LV relaxation pattern seen on echocardiography was associated with the metabolic syndrome in subjects younger than 65 years of age, but not in those ≥65 years. [141] In a Japanese study from 2009, in which subjects were stratified by age, only subjects in their 40s with diabetes showed signs of decreased E/A ratio and Em compared to healthy controls; no difference being seen for those in their 50s or older.[142] However, Em was reduced in all age groups (from 40s to 80s) when hypertension was also present. In the study presented in Paper I, neither conventional Doppler echocardiography nor TDI measures of LV diastolic function differed significantly between glucometabolic groups in the AGES-RS cohort. Some associations were observed between measures of LV diastolic function and continuous glycaemia measures, but only in age-adjusted models. In the MPP-RES cohort, a significant trend of increasing Am, transmitral A and transmitral E, and decreasing Em/Am with increased glucometabolic impairment was observed (in groups and with continuous measures). However, in the post hoc analysis of middle-aged and elderly subjects, this trend was only significant in the middle-aged group.

The situation is similar for LVH. An association between glucometabolic impairment and LVH has been well documented in young and middle-aged diabetic and pre-diabetic subjects, with a stronger correlation among women [63, 84, 143, 144] However, it is still being debated whether this association is independent of BMI and hypertension.[145-147] As with LVDD, studies including elderly subjects (i.e. 70 years and older) are sparse. In the five studies listed in Table 18 three included measures of LVMI. In the study by Lee et al. LVMI increased with increasing glucometabolic impairment[127] while in the Japanese study by Masugata et al. no increase in LVMI among subjects with diabetes was observed in the upper age groups compared to healthy controls.[142] In the present work (Paper I) LVMI was positively associated with HbA1c in the AGES-RS cohort. In the MPP-RES cohort, a positive association was observed between LVMI and FPG in the elderly group (age-adjusted only), and an increase in LVMI was seen in the groups of subjects with FPG ≥5.0 mmol/l and impaired glucose metabolism compared to the reference group (both age groups). However, it is important to note that LVMI was remarkably low in the reference group. Only middle-aged subjects with IFG and new-onset diabetes had significantly increased LVMI compared to the second glucometabolic group, which also had normal FPG (5.1-5.5 mmol/l). Thus, in contrast to previous studies in young and middle-aged subjects, the trend towards increased LVH with poorer glucose metabolism did not seem to be as strong in elderly subjects in the present work.

Table 18. Studies on the association between glucometabolic impairment and LVDD or LVH including elderly subjects.

First author	Number of participants	Gender-stratified	Age (years)	TDI applied	Main findings		Limitations
					LVDD	LVH	
Andren, B.[125]	205 healthy controls 29 with diabetes	No (men only)	All aged 70	No	LA dimension increased in DM. No difference in E, A, E/A or IVRT	NA	No women
Lee, M.[127]	5201 categorized as NFG, IFG, new-onset diabetes or established diabetes	Yes	Range 65-103	No	Increasing E (men only) and A (both sexes) with increasing glucometabolic impairment	Increasing LVMI with increasing glucometabolic impairment (both sexes)	Large proportion of missing values (30-40%)
Goncalves, A.[141]	653 healthy subjects	No	Mean 61±10, divided at 65	No	Impaired relaxation pattern associated with the metabolic syndrome in subjects aged <65 years but not ≥65 years	NA	No gender stratification Only one combined measure of LVDD
Masugata, H.[146]	400, including healthy subjects and those with diabetes, HTN or diabetes+HTN	No	Stratified (40s,50s, 60s, 70s and 80s)	Yes	E/A and Em decreased in subjects with isolated diabetes in their 40s compared to Controls, but not in subjects in their 50s or older	LVMI increased in subjects with diabetes in their 40s compared to controls, but not in subjects in their 50s or older	No gender stratification Only TDI value measured was Em
Leosdottir, M.	608 (AGES-RS) 1602 (MPP-RES)	Yes	Mean 76±6 and 67±6, MPP-RES stratified at age 70	Yes	No consistent associations between glucose metabolism and LVDD in AGES-RS or elderly in MPP-RES. A and Am increased, E/A decreased with increasing glucometabolic impairment in middle-aged in MPP-RES.	Increasing LVMI with increasing glucometabolic impairment, stronger in women.	No OGTT Risk for survival bias

Based on the findings in the above-cited studies and those presented in Paper I it can be concluded that the correlation between glucometabolic impairment and LVDD and LVH is not as strong in elderly populations as in middle-aged or young populations.

Nt-proBNP

Previous studies on the association between natriuretic peptide levels and diabetes have shown somewhat inconsistent results. Magnusson et al. found diabetic subjects without overt CVD to have higher mean Nt-proBNP levels than their non-diabetic counterparts.[94] No gender stratification was applied in that study; just over half of the subjects were women. In the course of the Framingham Study, Wang et al. found lower BNP levels in diabetic than in non-diabetic subjects of both sexes, more so in women.[148] More recently, Wang et al. published a study on the association between NPs and metabolic risk factors in the same cohort, where BNP was negatively associated with FG level, non-significantly in men but highly significantly in women.[149] In the study described in Paper II a positive correlation was found between Nt-proBNP and FPG in middle-aged men, but a non-significant inverse relationship in middle-aged women. Likewise, middle-aged diabetic men were found to have higher mean Nt-proBNP levels and middle-aged diabetic women lower Nt-proBNP levels than the reference groups (FPG≤5.0 mmol/l). Among the elderly subjects, however, the correlation between FPG and Nt-proBNP was non-significant for elderly men when analysing the whole echocardiography subcohort, and was non-significant for the elderly women, and no significant differences in mean Nt-proBNP were seen between those with normal FPG and those with impaired glucose metabolism. The conclusion drawn from the findings in Paper I, i.e. that LVDD and LVH assessed by echocardiography are not more common in elderly subjects with impaired glucose metabolism than in those with FPG within the normal range, is thus strengthened by the results presented in Paper II.

The Framingham Study group was the first to show that the ability of BNP to detect LVSD and LVH (by means of receiver operating characteristic) in non-diabetic populations is poorer in women than in men.[150] This observation is in agreement with the present findings, that the variance in Nt-proBNP was less well explained by the variables used in women than in men (40.2% vs. 52.3%), and only approximately 11% of the Nt-proBNP variance among women was explained by variables measuring LVH, LVSD and LVDD, compared to 19% among men. One possible explanation of this gender difference may be the correlation between NPs and the proportion of body fat. Serum concentrations of NPs are inversely correlated to BMI.[151] The mechanisms suggested to be responsible for this are the presence of NP clearance receptors in fat tissue (BNP only), passive clearance in extra-renal tissues, and obesity-related suppression of synthesis and/or secretion.[152] Women in all the glucometabolic groups apart from the reference group had a higher BMI than the men in the corresponding groups (p=0.04-0.006). This might partly explain the gender difference observed in this cohort, strengthened by the fact that the negative correlation in women was considerably weakened after adjustment for BMI. In the Framingham Study, however,

the inverse correlation between FG and BNP remained even after adjustment for BMI.[149] This could be because BMI is not an optimal measure of percentage body fat. Waist circumference, body weight and waist-hip ratio, are not considered better markers of body fat and therefore, substituting BMI with these variables in the regression analyses did not alter the present results.

Another possible explanation could be the potential influence of confounders not adjusted for in this work. Even after considering age, kidney function and BMI – variables known to strongly influence Nt-proBNP values – only 40.2% and 52.3% of the variance in Nt-proBNP could be explained for women and men, respectively. Consequently, the majority of the variance remains unexplained and the gender differences observed could be the result of unknown confounders.

In order to try to explain the findings presented in Papers I and II the correlation between Nt-proBNP and echocardiographic variables reflecting LVSD, LVDD and LVH was examined in the MPP-RES echocardiography subcohort. Nt-proBNP was found to be strongly correlated with LA area/m^2, Am, E/Em and LVMI for diabetic and non-diabetic men and women in both age groups. Although the p-values for middle-aged diabetic women were the weakest, the correlation coefficients were similar in all groups and it is thus likely that the weaker p-values reflect the small size of the group and not the actual strength of the relationships. The results of previous studies have been inconsistent, some concurring with the present results [93, 102, 153, 154], while others have reported no or weak correlations between NP levels and echocardiographic measures of LV dysfunction in diabetic subjects.[98, 99, 103, 104] Taking the inconsistency of the available evidence into consideration, caution is probably warranted when interpreting Nt-proBNP values in diabetic subjects, especially in women.

The conclusions that can be drawn from Papers I and II and the complementary results is that while LVDD and LVH could be identified by the Nt-proBNP level, irrespective of age and diabetes status, LVDD and LVH did not seem to be more common among elderly subjects with impaired glucose metabolism than in those with NFG. Furthermore, the associations between Nt-proBNP and increasing glucometabolic impairment in the elderly group were inconsistent; increasing glucometabolic impairment being associated with lower Nt-proBNP values in women, while the association was positive in men. Caution should therefore probably be exercised when interpreting Nt-proBNP values in these populations.

Risk factors, morbidity and mortality

In the work presented in Paper III it was observed that CVD risk factor burden increased linearly with increasing glucometabolic impairment among men and women in both age groups. The slope of the trend curve was not significantly steeper for middle-aged than elderly subjects of either sex, indicating that there was no significant weakening of the relation between increasing glucometabolic impairment and CVD risk factor burden with age. However, elderly men with impaired glucose metabolism had significantly fewer risk factors than middle-aged men in the corresponding groups while no such difference was observed among women. Consequently, the slope of the trend curve was significantly steeper for elderly women than elderly men, indicating a greater risk factor accumulation with increasing glucometabolic impairment among the elderly women. Elderly subjects of both genders reported worse SRH than middle-aged subjects. This difference decreased with increasing glucometabolic impairment among men, but not among women.

On the other hand, increasing HRs for CVD events with increasing glucometabolic impairment were only observed among middle-aged men and women, while elderly men in the first four groups (FPG ≤5.0-6.9 mmol/l) and elderly women in the first three groups (FPG ≤5.0-6.0 mmol/l) had HRs for CVD events close to 1.0 (Paper IV). There was thus a weakening of the association between increased risk of CVD and increasing glucometabolic impairment with increasing age. Despite similar values reflecting the strength of the interaction test for men and women, the difference between age groups was significant only among men. This can probably be explained by a lack of statistical power, as there were fewer women with fewer events during follow-up in the MPP-RES cohort. It was therefore concluded that there was no gender difference in weakening of the association with increasing age.

The increased risk of CVD among subjects with diabetes is indisputable and extensively documented.[11, 72, 155] However, some studies have reported a reduction in the ratios of CVD prevalence, incidence and mortality with increasing age between diabetic and non-diabetic subjects. Nichols et al. reported a CVD prevalence ratio of 4.00 (2.20-7.02) between diabetic and non-diabetic subjects (no gender stratification) aged 45 years and younger, while the corresponding ratio for those over 85 years of age was 1.26 (1.13-1.39).[156] A Norwegian study on CHD mortality showed a decline in risk ratio from the youngest (<60 years) to the oldest (>80 years) age groups, for both men and women.[123] In the Nurses' Health Study, however, only a minimal difference in CHD mortality risk ratios was found between women with and without diabetes in the age ranges 55-64 vs. >65 years.[157] In the NHANES survey from 1999, CHD mortality risk ratios between diabetic and non-diabetic men halved between the age groups 55-64 years and 65-74 years, while they remained unchanged for women.[158] Several studies have been carried out on FPG as a predictor of CVD events, including mainly elderly individuals. In the Cardiovascular Health Study by Smith et al. non-diabetic subjects (≥65 years, mean 73 years) were divided into

quintiles according to FPG, and CVD event prediction was evaluated.[159] Only the highest quintile (FPG >6.1 mmol/l) was predictive of CVD events. Gender-specific results were not reported. In the Dubbo Study in Australia, it was found that FPG levels were predictive of CHD incidence in women but not in men in non-diabetic subjects ≥60 years (mean age approximately 69 years).[160] Significance was, however, lost when excluding those with prevalent CVD, and in a complementary analysis of FPG quartiles only the fourth quartile significantly predicted CHD events. The Northern Manhattan Study included elderly individuals (mean age 68.8 years) of different ethnicities.[161] FPG was found to predict vascular events in the Hispanic and African-American subcohorts but not in the Caucasian subcohort. However, the mean age in the Caucasian subcohort was 73.4 years, compared to 65.5 and 71.6 years in the Hispanic and African-American subcohorts, respectively. The age-span in all these studies was quite wide and age stratification was not applied in any of them.

In the current work (Paper III), the risk factor burden increased significantly with glucometabolic impairment for men and women in both age groups. Although there were significant differences in risk factor burden between the middle-aged and elderly groups of men with impaired glucose metabolism, the difference in trends for risk factor accumulation across glucometabolic groups between the two age groups was not significant for either sex. This discrepancy might reflect a survival bias, in that subjects with impaired glucose metabolism experience a lifelong tendency for risk factor clustering, while those with the most serious metabolic impairment succumb to CVD events at an earlier age. This conclusion is supported by the studies cited above, as well as the observations reported in Paper IV, where there was an evident weakening of the predictive effect of FPG with age for both men and women.

However, survival bias is not the only possible explanation of these observations. FPG has been reported to increase physiologically with age.[162] In an interesting study by Yashin et al., using data from the Framingham cohort, a Cox model was applied to project all-cause mortality risk for blood glucose at given ages.[163] Apart from an increase in blood glucose with age, they showed that the range of deviations from the norm giving minimal increases in the relative risk of death also increased with age (Figure 10). FPG values in the upper range, which are considered pathological and convey an increased relative risk of death among subjects at younger ages, may thus be in the physiologically normal range for an elderly individual.

Finally, low FPG values can be a sign of underlying morbidity, especially among elderly subjects.[155, 164] The results presented in Paper IV give support to this hypothesis, as elderly men and women in the second (5.1-5.5 mmol/l) and third (5.6-6.0 mmol/l) glucometabolic groups had HRs below 1.0 (using subjects with FPG <5.0 mmol/l as the reference group).

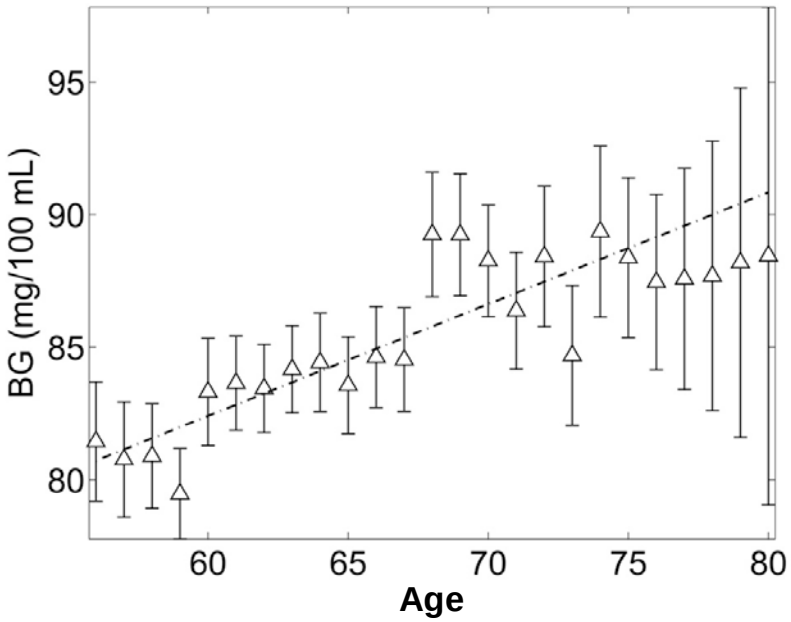

Figure 10. Normal blood glucose for females and males in the Framingham Study estimated using a semiparametric extended Cox-type model. Reprinted from Yashin et al.[163] with permission from Elsevier. BG, blood glucose.

In light of the age-related changes in what is regarded as NFG it might appear incongruous that IFG is classified as a FPG of 6.1-6.9 mmol/l according to the WHO[29] and 5.6-6.9 mmol/l according to the ADA[38] without any consideration of age. The argument for the chosen cut-off has been that IFG in general conveys an increased risk of progression to type-2 diabetes and possibly also the development of CVD. While the latter can be supported by the present results from the middle-aged subgroup, the same cannot be stated for the elderly subgroup, where the increased HRs for CVD events were only evident in those with new-onset and established diabetes.

Diabetes – an accelerator for physiological ageing?

Bearing in mind the relationship between age, diabetes and CVD, impaired glucose metabolism can be regarded as an accelerator for physiological ageing. Interestingly, the changes in LV diastolic function and structure seen in young and middle-aged diabetic subjects are similar to physiological age-related LV changes. Apart from LVDD, ageing is associated with increasing LV mass and LA diameter, and with decreasing longitudinal LV systolic function (measured by TDI), whereas LVEF is usually preserved.[165-167] Similarly, aortic stiffness and increased pulse pressure, well known components of vascular ageing, have been related to impaired glucose metabolism[168] In a study by Stacey et al. a decrease in aortic distensibility was observed in subjects with diabetes and IFG under 65 years of age, but impaired glucose metabolism was not a predictor for decreased aortic distensibility in those aged 65 years and older.[169]

Cardiovascular changes resulting from ageing and diabetes share not only physiological similarities but also biological ones. Decreasing telomere length has been linked to senescence.[170] Telomere attrition has also been shown to be associated with insulin resistance[171] and type 2 diabetes[172] and has been suggested as a biomarker of premature cellular aging in vascular and metabolic disorders.

Recently, a revision of Dzau's model of the so-called Cardiovascular Continuum was published, introducing a vascular ageing component to the original model (Figure 11).[173-175] The new model describes well the interaction between the pathological development of CVD, caused by unfavourable risk factors such as diabetes, and the "physiological" process of myocardial and vascular ageing. Both share a common final pathway of myocardial ischaemia, causing cardiac remodelling, and LV diastolic and systolic dysfunction, which finally lead to end-stage heart disease with symptomatic HF and death. If the common path is approached from the left (see Figure 11), as in the case of subjects with early or midlife onset diabetes, hypertension or the metabolic syndrome, subjects will enter the final stages of the continuum at an earlier age than their peers. As a consequence, they should be easily identified from the healthier mass by means of, for instance, echocardiography and mortality risk projections. Indeed, Masugata et al. demonstrated that patients with diabetes in their 40s and 50s had signs of LVSD and LVDD comparable to those seen in healthy subjects 30 years older.[142]

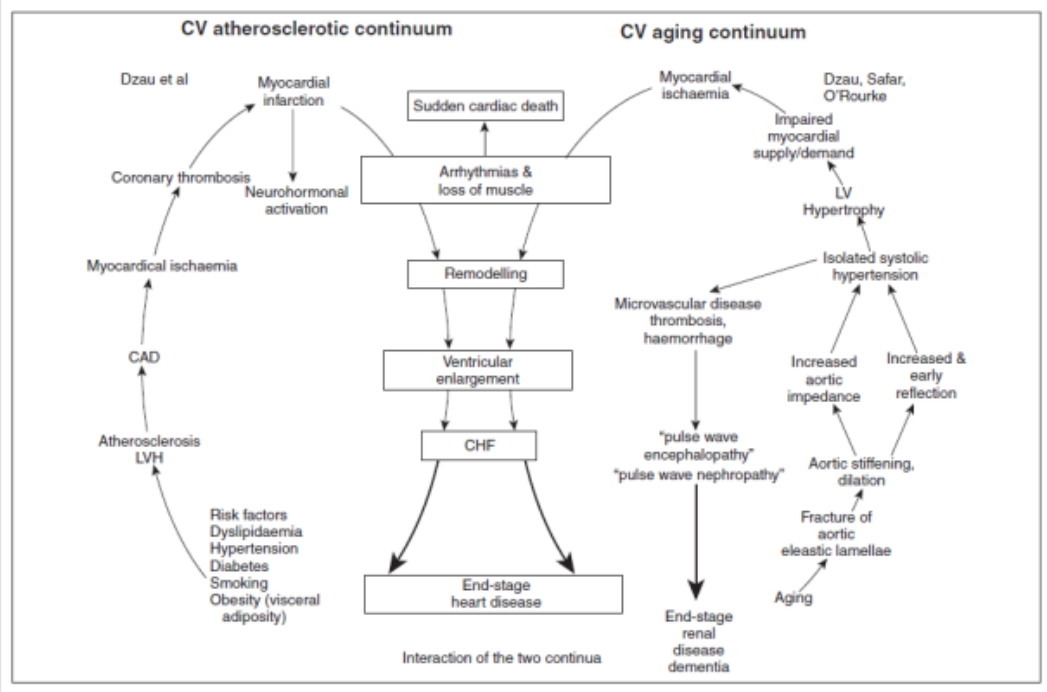

Figure 11. The classic Cardiovascular Continuum (left) and the Aging Continuum (right), and their interaction. Reprinted from O'Rourke et al.[175] with permission from Sage Publications. CV, cardiovascular; CAD, coronary artery disease; CHF, congestive heart failure.

An individual who acquires diabetes at an older age, however, may be approaching the common stages at a similar time to his or hers peers, as both are already experiencing LV remodelling and signs of end-stage heart disease solely as a result of advancing age. Consequently, and a likely explanation of the findings presented in Papers I, II and IV, age-related changes in LV structure and function dominate the changes in LV diastolic function expected with impaired glucose metabolism. It can also be speculated that those with unfavourable metabolic profiles in midlife will already have died at an earlier age, and are thus not represented in cohort studies such as MPP-RES and AGES-RS, i.e. there is selection bias.

Gender issues

Although recent international studies still indicate that women bear a disproportionate part of the diabetes-related excess CVD risk[111, 155] the difference between the sexes seems to have declined since it was first described in the Framingham study in the late 1970s, when Kannel and colleagues reported an 3.6-fold increase in the risk of MI for diabetic women and a 1.8-fold increase in risk for diabetic men, compared to their non-diabetic counterparts.[176] In Sweden, the excess risk of mortality for diabetic compared to non-diabetic subjects has declined for women over the recent decades, while remaining the same for men.[177] Also, it was found in a recent Swedish study that the excess CVD mortality risk for diabetic subjects was smaller than in other studies (1.41 (1.17-1.70) for women and 1.27 (1.06-1.52) for men).[178] There have been speculations as to the reasons for this difference. In the INTERHEART study, diabetes contributed more to the population-attributable risk of MI in women than men.[6] Consistent with the findings presented in Paper I, a stronger association has been reported between impaired glucose metabolism and LVMI for women than for men.[144] Disparities in the diagnosis and treatment of CVD, as well as gender-related differences in response to treatment have also been suggested as explanations.[117] It has also been suggested that there is a higher CVD risk factor burden in diabetic women than in diabetic men.[113] The results presented in Paper III support this; the women in all glucometabolic groups in both age groups exhibited more CVD risk factors than men. The same trend was observed for SRH, in itself a strong predictor of all-cause and disease-specific mortality, also among subjects with diabetes.[106, 107, 179] The HRs for CVD event risk presented in Paper IV were higher for women with new-onset diabetes in both age groups than in men, although the statistical significance was borderline. There were minimal differences in HRs between men and women with IFG or established diabetes, and the differences were weaker for all three groups with impaired glucose metabolism in the fully adjusted models. In conclusion, our results concur with previous reports on decreasing gender differences in diabetes-related excess risk for CVD, but still support the fact that not only women with diabetes but also pre-diabetic women, irrespective of age, should be given special attention when screening for and treating concomitant CVD risk factors.

Limitations

The studies described in this thesis have several limitations, which are discussed below.

Healthy cohort effect

As with all population-based cohort studies there is risk of a "healthy cohort" effect, or survival bias. In other words, subjects with co-morbidities, disability and poor quality of life might either have died or been too weak to participate in the study. As these subjects would be more likely to be older and classified in the groups of subjects with greater glucometabolic impairment, such a bias would lead to underestimation of any cardiac risk related to glucometabolic disturbance in that group. A healthy cohort effect might be reflected in the average HbA1c levels of the subjects with impaired glucose metabolism in both the MPP-RES and AGES-RS cohorts, which were <6.0% for those with new-onset diabetes in both studies and approximately 6.5% for those with established diabetes in the MPP-RES cohort and 5.5% in the AGES-RS cohort. Also, a study on participants versus non-participants was performed in the original Malmö Preventive Project, showing that total and cause-specific mortality were higher among non-participants than in those participating in the study.[66] It can be assumed that similar results would be found in the MPP-RES, and indeed, in all population-based studies including elderly subjects.

Applicability and cohort size

The MPP-RES and AGES-RS cohorts were recruited from single urban study centres, limiting the applicability of the results to other populations. However, the reproducibility of the results from the two independent cohorts strengthens the validity of the findings, albeit negative (Paper I).

There were fewer women than men in the MPP-RES cohort, resulting in limited numbers of women in the various glucometabolic groups. This was especially noticeable in the echocardiography subcohort (Papers I and II) and the number of CVD events among women with impaired glucose metabolism (Paper IV). This could have reduced the power to detect small differences between groups of women.

Confounders

Many of the subjects were taking prescription medication affecting cardiac function. To minimize confounding effects subjects with prevalent CHD, HF and valvular disease were either excluded from all (Paper I) or repeated (Papers II) calculations, adjustments were made for prevalent CVD and HF (Paper IV) or adjustments were made for medication (Papers I, II and IV). However, the effect of medication can probably never be fully accounted for and might have biased the results, especially as those with greater glucometabolic impairment were more heavily medicated. However, a study population consisting of elderly individuals from which all those on

prescription medication for CVD or hypertension have been excluded would not be representative of an elderly background population.

Measurements and classification of impaired glucose metabolism

No OGTTs were performed in either the MPP-RES or the AGES-RS. Also, it cannot be ruled out that some glucose values were non-fasting. Additionally, only one measurement of FG was performed in the AGES-RS. This might have led to misclassification of glucometabolic impairment and underestimation of glucometabolic-related risk, especially in women, as IGT and diabetes defined by an OGTT is more common among women than men.[72] Also, as IGT is more common and a better predictor of CVD than IFG in older age 2-hour glucose would probably have been a better indicator of glucose metabolism among the elderly than FG.[180] Additionally, both the duration and severity of diabetes influence diabetes-related myocardial dysfunction.[157, 181] The duration of disease was included in the self-administered questionnaire in the MPP-RES but the results were unreliable due to a large proportion of missing data, and this was thus not included as a confounder in the current analyses. In the case of an average short duration of diabetes this might have biased the results, increasing the likelihood of negative findings. This must be deemed plausible in light of the likelihood of a healthy cohort effect and low mean HbA1c values among subjects with impaired glucose metabolism.

Study design and statistical considerations

The studies presented in Papers I-III are of cross-sectional design, so no conclusions can be drawn regarding causality. Considering risk factors for CVD, rather than CVD morbidity and mortality (Paper III) limits the conclusions that can be drawn about the clinical relevance of the results. However, by including CVD events (Paper IV) the questions raised in Paper III could at least be partially answered. Unfortunately, the statistical power was sometimes not sufficient to reveal significant differences in Paper IV, especially among women, as there were few CVD events in the smaller groups (e.g. those with new-onset diabetes). The intention is to collect endpoint data for the rest of 2009 and the whole of 2010 (due in August 2012) and thereafter repeat the calculations. This manuscript will not be submitted for publication until after this revision.

Finally, due to multiple testing (especially in Paper I) borderline p-values should be interpreted with caution.

CONCLUSIONS

The main conclusions that can be drawn from the studies described in this thesis are presented below.

- LVDD, assessed by tissue Doppler imaging, is not more common in elderly men or women with impaired glucose metabolism than in those with fasting glucose levels within the normal range (Paper I).

- Although the degree of explanation of the variance in Nt-proBNP by echocardiographic measures of LVH, LVDD and LVSD among middle-aged and elderly subjects seems relatively low, Nt-proBNP is still strongly correlated with measures of LVDD in both age groups, irrespective of diabetes status. However, in agreement with the findings reported in Paper I, correlations between Nt-proBNP and FPG are weak and inconsistent among elderly subjects (Paper II).

- The association between risk factor accumulation and poor SRH with increasing glucometabolic impairment is not weakened with advancing age, being strong for both middle-aged and elderly subjects of both sexes. Risk factor accumulation seems relatively more pronounced in elderly women than in elderly men (Paper III).

- The relation between CVD events and glucometabolic impairment does seem to become weaker with advancing age, as evidenced by FPG values within the upper pre-diabetic range conveying less increased risk of CVD events in elderly than in middle-aged men and women (Paper IV).

- Generally, women with impaired glucose metabolism have a higher LVMI, heavier risk factor burden, poorer SRH and relatively higher HR for CVD events than the corresponding men, although the gender difference seems less pronounced than previously reported.

FUTURE PERSPECTIVES

The studies included this thesis have described the relation between glucose metabolism and aspects of CVD – namely signs of myocardial dysfunction, risk factor burden and incident CVD events – focusing on elderly subjects. While answering some questions, new ones have also been raised.

The results imply that LVDD is not more common in elderly subjects with impaired glucose metabolism than in those with NFG. Also, even though raised FPG levels below the diabetes threshold were correlated with an increasing risk factor burden they did not seem to convey an increased risk for CVD. The fact that poor SRH was correlated with increasing FPG might, however, imply that elderly subjects with impaired glucose metabolism experience complications other than CVD, such as microvascular complications or other metabolic perturbations increasing the risk of progression to diabetes, neither of which were studied in the work presented in this thesis. Also, while FPG was not a predictor of CVD events it was an indicator for the presence of other CVD risk factors, which in turn might predict the future risk of disease and disability. All of these areas need to be studied further in elderly populations.

Epidemiological studies are the preferred study model to describe correlations, identify risk factors and project future event risks in defined populations. However, correlations do not imply causation and cohort studies are by definition only descriptive. Intervention trials are needed to establish whether relationships described in epidemiological studies can be influenced and risks reversed. Results from large-scale intervention trials have raised questions regarding the benefit of intensive glucose-lowering treatment among subjects with type 2 diabetes, especially among elderly subjects and those with a long duration of disease.[182] Also, treating pre-diabetic subjects to prevent future CVD has not proved as positive as once hoped for.[183, 184] The effect of concomitant risk factor treatment and lifestyle intervention has been more promising, with supporting evidence even in elderly populations [185, 186] More studies are nevertheless needed and these need to include elderly subjects and report on gender-stratified results. Also, new medications with different mechanisms of action are continuously being introduced, so the quest for better treatment is far from over.

With the projected changes in the age composition of the world's population, the prevalence of CVD and its complications will continue to increase. The same is true of impaired glucose metabolism, at least until lifestyles are changed towards more healthy living. The personal disability and cost to society caused by these diseases are enormous. While treating established disease is important, the need for enforced prevention strategies is all the more called for.[12, 187] The resources available for screening and prevention are unfortunately minimal compared to those spent on

treatment and clinical care. With the majority of the population-attributable risk being modifiable this needs to change. Upstream thinking, with emphasis on healthy living from an early age, should be a top priority for societies and health care systems alike.

Finally, the results presented in this thesis clearly show that caution should be applied when generalizing evidence from studies in other age groups to the elderly, and from non-gender stratified studies to what is relevant for men and women.

SUMMARY IN SWEDISH

Studier har visat att inte bara individer med diabetes utan också de som har förstadier till diabetes, nämligen förhöjt fasteblodsocker och nedsatt glukostolerans, löper en ökad risk för att drabbas av såväl hjärtkärlsjukdom som hjärtsvikt. Däremot har man sett att den ökade risken förenad med diabetes verkar minska med åldern. Studier på äldre individer är dock sparsamma, de studier som finns är begränsade till diabetiker, och ofta rapporteras inte resultaten åtskilt för män och kvinnor. Det övergripande syftet med de fyra delarbeten som presenterats i denna avhandling var att ta reda på om det finns skillnader mellan medelålders och äldre män respektive kvinnor avseende sambandet mellan störd glukosmetabolism (från normalt fasteblodsocker till känd diabetes), riskfaktorer för hjärtkärlsjukdom, tidig hjärtdysfunktion och risken för hjärtkärl-händelser såsom hjärtinfarkt och stroke.

På 1970-talet startade ett primärpreventivt projekt vid Medicinska kliniken, UMAS (numera Skånes Universitetssjukhus) i Malmö. Projektet som kallades Malmö Förebyggande Medicin (MFM) började 1974. Under 17 år blev totalt 33 000 män och kvinnor undersökta med frågeformulär, blodprover och kroppslig undersökning. I början av 2000-talet bjöds alla kvarlevande som deltagit i MFM in till en återundersökning där återigen deltagarna fick svara på frågeformulär och fick taget blodprov, man tog puls och blodtryck, mätte längd, vikt, stuss och midjemått. Totalt deltog drygt 18 000 medelålders och äldre män och kvinnor denna gång. Hos en mindre grupp (ca 1800 deltagare) genomfördes ultraljudsundersökning på hjärtat, man tog EKG och ett speciellt blodprov som kallas Nt-proBNP, vilket återspeglar belastning i hjärtat. Deltagarna har sedan dess följts upp i nationella register avseende hjärtkärl-händelser.

Ett liknande projekt, Reykjavik studien, genomfördes i Island på sjuttio- och åttiotalet. Även där kallades deltagarna tillbaka i början på 2000-talet. Totalt deltog ca 5700 i återundersökningen i Reykjavik, varav 900 undersöktes med ultraljud på hjärtat. Dessa två stora befolkningsstudier gav underlag till studierna i avhandlingen.

Bland hjärtfriska medelålders (57-69 år) män och kvinnor blev hjärtdysfunktion mätt med ultraljud mer uttalad ju högre socker deltagarna hade. Samma fynd såg man inte bland äldre (70-80 år) män och kvinnor där de som hade sockerrubbningar inte hade tecken till hjärtdysfunktion i någon större utstreckning än de som hade normal sockerbalans.

Hos medelålders som män steg Nt-proBNP jo högre socker männen hade. Ett positivt samband mellan socker och Nt-proBNP sågs också hos de äldre männen, men där var sambandet svagare och icke statistiskt signifikant. Hos kvinnorna sågs ett omvänt samband – jo högre socker de hade, desto lägre Nt-proBNP värde. Sambandet var dock varken signifikant hos de medelålders eller de äldre kvinnorna. Fynden

förstärker de från ultraljudsundersökningarna, d.v.s. att hjärtdysfunktion verkar inte förekomma i större utstreckning hos äldre individer med sockerrubbningar än hos de som har normalt socker.

När man analyserade riskfaktorer för hjärtkärlsjukdom (såsom högt blodtryck, rökning och högt kolesterol) och självskattad hälsa hittade man däremot en klart ökad riskfaktorbelastning och sämre självskattad hälsa med stigande blodsocker både hos medelålders och äldre deltagare. Över lag så var riskfaktorbördan större och den självskattade hälsan sämre hos kvinnor, och dessa försämrades mer med stigande socker bland de äldre kvinnorna än de äldre männen.

När man till slut analyserade hjärtkärlhändelser såg man att ju högre socker medelålders män och kvinnor hade vid studiestarten, desto större risk fanns för att de hade drabbats av hjärtinfarkt eller stroke under uppföljningstiden. Samma ökning sågs inte bland de äldre, där sockret verkade inte ha någon större effekt på risken. Hos både medelålders och äldre män och kvinnor med diagnosticerad diabetes, däremot, var risken för att utveckla hjärtinfarkt eller stroke ökad jämfört med de som hade normalt fasteblodsocker.

Slutsatserna att dra av dessa studier är att förhöjt blodsocker inte verkar ha lika mycket effekt på hjärtfunktionen och risken för att drabbas av hjärtinfarkt och stroke hos äldre som hos medelålders män och kvinnor. Att ha förhöjt blodsocker är dock förenad med en ansamling av övriga riskfaktorer för hjärtkärlsjukdom och försämrad självskattad hälsa hos äldre likaväl som hos de som är i medelåldern, mer så hos kvinnor än hos män relativt sett.

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