Associations of plasma renin with 10-year cardiovascular mortality, sudden cardiac death, and death due to heart failure

Andreas Tomaschitz1*,†, Stefan Pilz1,2†, Eberhard Ritz3, Alberto Morganti4, Tanja Grammer5, Karin Amrein1, Bernhard O. Boehm6, and Winfried März5,7,8

1Department of Internal Medicine, Division of Endocrinology and Metabolism, Medical University of Graz, Auenbruggerplatz 15, A-8036 Graz, Austria; 2Department of Epidemiology and Biostatistics and EMGO Institute for Health and Care Research, VU University Medical Center, Amsterdam, the Netherlands; 3Department of Nephrology, University of Heidelberg, Heidelberg, Germany; 4Department of Internal Medicine and Hypertension Center, San Giuseppe Hospital, University of Milan, Milan, Italy; 5Synlab Centre of Laboratory Diagnostics, Heidelberg, Germany; 6Division of Endocrinology, Diabetes and Metabolism, Graduate School of Molecular Diabetology and Endocrinology, Ulm University, Ulm, Germany; 7Clinical Institute of Medical and Chemical Laboratory Diagnostics, Medical University of Graz, Graz, Austria; and 8Mannheim Institute of Public Health, Rupertus Carola University Heidelberg, Medical Faculty Mannheim, Mannheim, Germany

Received 13 December 2010; revised 15 March 2011; accepted 16 April 2011; online publish-ahead-of-print 23 May 2011

See page 2610 for the editorial comment on this article (doi:10.1093/eurheartj/ehr187)

Aims
Renin is the key regulator of the renin—angiotensin—aldosterone system. Previous studies have reported conflicting results on the relation of plasma renin with fatal cardiovascular events. This study in a large cohort of patients sought to evaluate the association between plasma renin concentration (PRC) and cardiovascular mortality after long-term follow-up of almost 10 years.

Methods and results
Plasma renin concentration [median: 11.4 (6.0–24.6) pg/mL] was measured in 3303 patients (mean age: 62.7 ± 10.6 years; 30.3% women) referred to coronary angiography. After a median follow-up of 9.9 years, 554 participants (16.8%) with PRC measurement at baseline had died due to fatal cardiovascular events. Multivariable-adjusted Cox analysis revealed that when compared with participants in the lowest PRC quartile, those in the highest quartile were at increased risk of cardiovascular mortality (hazard ratio: 1.79, 95% CI 1.28–2.48). Analyses of specific causes of cardiovascular death showed that for each standard deviation increase in log-PRC there was a 22% (P = 0.006) increase in risk of sudden cardiac death and a 23% (P = 0.033) greater risk of death due to heart failure. The association of PRC with cardiovascular mortality remained stable after adjustment for established cardiovascular risk factors, ongoing antihypertensive medication, immunoreactive angiotensin II, and aldosterone levels. Age, N-terminal pro-B-type natriuretic peptide levels, coronary artery disease, the use of angiotensin-converting enzyme-inhibitors, beta-blockers, diuretics, and kidney function were important effect modifiers.

Conclusions
Plasma renin concentration is associated with long-term cardiovascular mortality in patients referred to coronary angiography. Further intervention studies should determine whether renin is a potential therapeutic target or only a marker of mortality risk in various cardiovascular risk groups.

Keywords
Plasma renin • Cardiovascular mortality

Introduction
Activation of the renin—angiotensin—aldosterone system (RAAS) molecular cascade maintains circulatory homeostasis in the human body.1 Continued and inappropriate activation of this system, i.e. in the course of chronic heart failure (CHF), renal dysfunction, or acute myocardial infarction (MI), might contribute to the development and progression of cardiovascular diseases (CVDs).2–5 So far, potential tissue damaging properties of inappropriately activated circulating and tissue RAASs have been mainly attributed

* Corresponding author. Tel: +43 316 385 12383; fax: +43 316 385 13428. Email: andreas.tomaschitz@gmx.at
† Both authors contributed equally to this work.
Published on behalf of the European Society of Cardiology. All rights reserved. © The Author 2011. For permissions please email: journals.permissions@oup.com
to angiotensin II (Ang II). Accumulating evidence however points to renin, which catalyses the rate-limiting step of the RAAS by converting angiotensinogen to Ang I, as an important mediator of vascular and myocardial injury. Renin mediates an increased formation of Ang II and aldosterone and activates an array of intracellular signalling pathways in cardiomyocytes, vascular and renal cells by binding to the (pro)renin receptor (PRR).6

Previous studies that investigated the association between renin and CV damage reported inconsistent findings. In 1973, Doyle et al.7 found no relation between plasma renin and the risk of vascular complications in hypertensive patients. In line with this, Meade et al.8 found no significant relation between plasma renin and higher risk of MI and sudden cardiac death (SCD) in normotensive men. In contrast, Alderman et al.9 reported an independent association between plasma renin and risk of MI in hypertensive patients. In the Framingham Offspring Study, plasma renin was significantly related to higher risk of death, whereas in the same cohort Parikh et al. could not find a significant association between plasma renin and CVD mortality after 3 years of follow-up.10,11

Most of the studies that investigated the relation between plasma renin and CV events were limited by low sample size, laboratory methods of renin measurement, short-term follow-up, small numbers of outcome events, and inappropriate consideration of confounding CV risk factors and renin-modulating factors, i.e. antihypertensive medication. Moreover, because of the prevalence of Ang II as well as aldosterone levels on plasma renin-related cardiovascular damage have not been considered in previous analyses, data regarding the association between renin and CVD mortality remain a matter of debate.

The aim of the present prospective analysis was to evaluate the impact of a plasma renin on the risk of CVD mortality in a large cohort of patients referred to coronary angiography. We analysed the relation between plasma renin and risk of CV death under consideration of important renin-modulating factors and after a sufficient long-term follow-up of almost 10 years.

**Methods**

**Study design and participants**

The Ludwigshafen Risk and Cardiovascular Health (LURIC) Study is an ongoing prospective cohort trial designed to investigate the effects of genetic polymorphisms and several biomarkers on the CV system. Study design and baseline examinations have been described previously in detail.12 In brief, between 1997 and 2000, 3316 Caucasian study participants without major non-CV diseases were referred for coronary angiography to the Department of Cardiology at the Ludwigshafen General Hospital, which is a tertiary centre, and enrolled in the study. Inclusion criteria were availability of coronary angiogram and stable clinical condition, except for the presence of an acute coronary syndrome (ACS). Patients, who suffered from any severe non-cardiac disease, were excluded. The LURIC study was approved by the institutional review board at the ‘Ärztekammer Rheinland-Pfalz’. Each participant provided written informed consent and our study complies with the Declaration of Helsinki.13

Coronary artery disease (CAD) was evaluated by coronary angiography based on maximal luminal narrowing of visual stenosis and defined as the presence of at least one stenosis ≥10% in at least one of 15 coronary segments of the three major coronary arteries. An ACS was diagnosed if patients presented within 7 days of onset of symptoms of unstable angina pectoris or acute MI, comprising non-ST-elevation MI (troponin T > 0.1 μg/L) and ST-elevation MI (troponin T > 0.1 μg/L). Previous MI was assumed based on a documented history of electrocardiographic ST- and non-ST-elevation and/or elevation of cardiac biomarkers. Brachial artery pressure measurements were taken with an automated oscillometric device (Omron M4X, Omron Health Care GmbH, Hamburg, Germany) after the patient had rested in the supine position for 10 min. At least three consecutive systolic and diastolic blood pressure (BP) measurements were taken with a minimum interval of 30 s. The severity of heart failure (HF) was assessed using the New York Heart Association (NYHA) classification and in 1360 (41.0%) participants left ventricular ejection fraction (LVEF) was additionally calculated from the right anterior oblique view. Dyslipidaemia was defined according to the recommendations of the National Cholesterol Education Program Adult Treatment Panel III as HDL cholesterol <1 mmol/L (40 mg/dL), and/or LDL cholesterol > 4.1 mmol/L (160 mg/dL), and/or triglycerides > 2.4 mmol/L (200 mg/dL). Diabetes was classified according to the American Diabetes Association including the new criterion of glycylated haemoglobin A1c (HbA1c) ≥6.5%. Finally, current smoking status and antihypertensive treatment [angiotensin-converting enzyme (ACE)-inhibitors (ACEI), Ang-II type-1 receptor blockers (ARBs), beta-blockers, calcium channel blockers, and diuretics] were recorded. All patients were on an unrestricted (Western) diet.

**Laboratory analysis**

Following an overnight fast, venous blood was sampled in the morning before coronary angiography between 6.10 a.m. and 10.30 a.m. (mean sampling time 7.22 a.m. ± 0.32 min), with the participants in supine position for 5–10 min before phlebotomy. Routine laboratory parameters were immediately measured whereas remaining blood samples were snap frozen for further determination and stored at −80 °C until analysis. The standard laboratory methods have been described previously in detail.12

Plasma renin concentration (PRC) was determined by immunoradiometric assay (Active Renin, Nichols Institute Diagnostics, San Juan, Capistrano, CA, USA) in 3303 participants. The conversion factor for PRC is based on a publication by Trenkel et al.14 1.67 μU/mL (=mU/L) renin equals 1 pg/mL (=ng/L) renin. The normal range in supine position is given as 3–28 pg/mL. Immunoassays measuring PRC are limited with regard to detection accuracy in the low renin range (<4 mU/L).15 However, PRC assays have a lower coefficient of variation compared with PRA assays for plasma samples with typical or increased values.15 No patients with impaired liver function (reflected by 2.5 times higher serum alanin transaminase and aspartate transaminase concentrations) have been enrolled in the LURIC study. This is important for our work when considering that liver damage is associated with reduced angiotensinogen synthesis and consequently reduced Ang II production. This in turn results in higher PRC values due to less feedback inhibition by Ang II resulting in constant Ang II levels.16 Plasma aldosterone concentration (PAC, pg/mL; conversion to pmol/L: ng/L × 2.78) was measured by radioimmunoassay (Active aldosterone, Diagnostic Systems Laboratories, Sinzheim, Germany). Overall correlation between this RIA and other commercial assays ranges between 0.74 and 0.98.17 The intra-assay and inter-assay coefficients of variation of this assay were 3.6–8.3% and 7.3–10.4%, respectively. The reference interval is given as 30–160 pg/mL. Immunoreactive Ang II (ir-Ang II, ng/L) was determined by an RIA
Follow-up
Information on vital status was continuously obtained from local person registries. Classification of death from CVD and non-CVD causes was based on detailed independent exploration of death certificates and medical records by two experienced clinicians who were blinded to the study data except for death certificates. Cardiovascular disease deaths were further categorized as SCD, fatal MI, death due to congestive HF, death immediately after intervention to treat CAD, and other causes of deaths due to cardiac disease. Sudden cardiac death was defined as a sudden unexpected death either within 1 h of symptom onset or within 24 h of having been observed alive and asymptomatic.15 Non-CVD deaths were classified into death from fatal infection, fatal cancer, and other (undefined) causes of death. In the case of disagreement about classification, the final decision was made by one of the principal LURIC investigators after appropriate review of the data.

Statistical analysis
Patients were divided into quartiles based on the distribution of PRC at baseline. Normally distributed continuous variables were given as mean [with standard deviation (SD)], variables with skewed distribution as median with interquartile range, and categorical variables as percentages. For parametric procedures all skewed distributed continuous variables were logarithmically transformed (log10). Baseline characteristics were compared across PRC quartiles using the χ² trend test with P by linear-for-linear test for categorical variables and by using an analysis of variance with P for trend for continuous variables. Hazard ratios (HR) and 95% confidence intervals (CI) for all-cause and CVD mortality were calculated using Cox proportional hazard analyses (with backward procedures) to estimate relative risk of incident fatal CVD events by baseline PRC categories (using the first quartile as reference). For PRC as continuous variable, the HR (with 95% CI) was calculated per increment of log SD (log10). Besides presenting a crude model, variables considered for multivariate model 1 included age, gender and for model 2 systolic BP, HDL- and LDL-cholesterol, ex and active smokers, CAD, body mass index, hsCRP, plasma aldosterone and ir-Ang II levels did not materially change (95% CI 0.86–1.39, P = 0.476), and 1.20 (95% CI 0.95–1.52, P = 0.118) compared with the lowest quartile.

Unadjusted Cox proportional HR for CVD mortality in the first vs. the second, third, and fourth quartile of PRC had a HR for death of 1.02 (95% CI 0.80–1.30, P = 0.874), 1.09 (95% CI 0.86–1.39, P = 0.476), and 1.20 (95% CI 0.95–1.52, P = 0.118) compared with the lowest quartile. Additional adjustments for CVD risk factors, detailed antihypertensive medication, plasma aldosterone and ir-Ang II levels did not materially change our results. Compared with patients in the first PRC quartile, the adjusted HRs for CVD death were 1.79 for patients in the fourth quartile (95% CI 1.28–2.48, P = 0.001). A multivariate-adjusted increase of 1 SD log-PRC was associated with a 20% higher risk of CVD mortality, indicating that lower PRC levels

Results
Baseline data
The mean age of 3303 study participants with PRC measurement at baseline [median PRC 11.4 (6.0–24.6) pg/mL] was 62.7 ± 10.6 years; 30.3% were female. In contrast to arterial hypertension, frequency of CAD, and severe HF (NYHA III/IV) were more frequently found in patients with higher PRC levels. The distribution of demographics, co-morbidities, and laboratory characteristics according to levels of PRC (in quartiles) is shown in Table 1.

Association of plasma renin with all-cause and cardiovascular mortality in the overall cohort
Eighteen individuals were lost during follow-up; in 24 study participants no death certificates were available and they were excluded from the mortality analyses. During a median follow-up of 9.9 years, 987 (29.9%) patients with available PRC measurement at baseline died. Of these, 618 patients (18.7%) died from CV and 369 (11.2%) from non-CV causes.

In 17 deceased patients we could not obtain sufficient information for the classification of the causes of death. These patients were included into analyses for all-cause mortality but were excluded from analyses of specific causes of death. Of the remaining 970 patients, 554 died due to CV causes including 258 SCD, 105 fatal MI, 146 deaths due to HF, 26 deaths after interventions to treat CAD, and 19 deaths due to other cardiac causes. Among non-CV causes, we recorded 61 fatal strokes, 143 fatal cancers, 76 fatal infections, and 134 other causes of death.
were related to higher survival probability. Assumptions underlying the Cox proportional hazard models i.e. the proportionality of hazards were evaluated using log-minus-log survival and partial (Schoenfeld) residuals vs. survival time plots and were found valid. For cardiovascular mortality, the C statistic for the fully adjusted Model 3 was 0.692 (95% CI 0.666–0.717) without PRC and 0.693 (95% CI 0.668–0.718) with PRC, indicating that the addition of PRC to establish CVD risk factors does not significantly improve prediction of CVD mortality. Kaplan–Meier analysis showed an increased probability of cardiovascular death during follow-up

### Table 1 Baseline characteristics according to plasma renin concentration quartiles

<table>
<thead>
<tr>
<th>PRC&lt;sup&gt;a&lt;/sup&gt; variable</th>
<th>Quartile 1 (&lt;5.5 pg/mL)</th>
<th>Quartile 2 (5.5–11.4 pg/mL)</th>
<th>Quartile 3 (11.5–24.0 pg/mL)</th>
<th>Quartile 4 (&gt;24.0 pg/mL)</th>
<th>P trend</th>
</tr>
</thead>
<tbody>
<tr>
<td>PRC (pg/mL)</td>
<td>n = 792</td>
<td>n = 889</td>
<td>n = 787</td>
<td>n = 835</td>
<td></td>
</tr>
<tr>
<td>3.6 (2.4–4.8)</td>
<td>3.6 (2.4–4.8)</td>
<td>3.6 (2.4–4.8)</td>
<td>3.6 (2.4–4.8)</td>
<td>3.6 (2.4–4.8)</td>
<td>–</td>
</tr>
<tr>
<td>Ir-Angiotensin II&lt;sup&gt;b&lt;/sup&gt; (ng/L)</td>
<td>13.0 (9.0–20.0)</td>
<td>16.0 (11.0–26.0)</td>
<td>21.0 (14.0–33.0)</td>
<td>35.0 (20.0–67.0)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>PAC&lt;sup&gt;c&lt;/sup&gt; (pg/mL)</td>
<td>67.0 (41.0–104.0)</td>
<td>69.0 (46.0–107.0)</td>
<td>84.0 (52.5–126.0)</td>
<td>100.0 (59.0–160.0)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Age (years)</td>
<td>63.4 ± 9.7</td>
<td>62.7 ± 10.5</td>
<td>62.2 ± 11.4</td>
<td>62.4 ± 10.9</td>
<td>0.048</td>
</tr>
<tr>
<td>Female sex (%)</td>
<td>40.9</td>
<td>30.9</td>
<td>25.7</td>
<td>24.3</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Body mass index (kg/m&lt;sup&gt;2&lt;/sup&gt;)</td>
<td>27.2 ± 3.9</td>
<td>27.4 ± 4.0</td>
<td>27.4 ± 3.9</td>
<td>27.9 ± 4.5</td>
<td>0.006</td>
</tr>
<tr>
<td>LDL cholesterol (mg/dL)</td>
<td>118 ± 32</td>
<td>117 ± 36</td>
<td>116 ± 35</td>
<td>115 ± 34</td>
<td>0.028</td>
</tr>
<tr>
<td>HDL cholesterol (mg/dL)</td>
<td>40 ± 11</td>
<td>39 ± 10</td>
<td>38 ± 11</td>
<td>38 ± 11</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Dyslipidaemia (%)</td>
<td>66.0</td>
<td>67.7</td>
<td>71.4</td>
<td>70.5</td>
<td>0.018</td>
</tr>
<tr>
<td>Current smokers (%)</td>
<td>17.3</td>
<td>19.0</td>
<td>21.1</td>
<td>21.7</td>
<td>0.015</td>
</tr>
<tr>
<td>Diabetes mellitus (%)</td>
<td>37.2</td>
<td>36.2</td>
<td>41.4</td>
<td>44.9</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>HbA1c (%)</td>
<td>5.9 (5.5–6.4)</td>
<td>6.0 (5.6–6.5)</td>
<td>6.0 (5.6–6.7)</td>
<td>6.1 (5.7–7.0)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>History of arterial hypertension (%)</td>
<td>61.7</td>
<td>56.9</td>
<td>57.4</td>
<td>58.8</td>
<td>0.310</td>
</tr>
<tr>
<td>Arterial hypertension&lt;sup&gt;d&lt;/sup&gt; (%)</td>
<td>79.0</td>
<td>73.3</td>
<td>71.5</td>
<td>67.7</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Peripheral systolic BP (mmHg)</td>
<td>147 ± 23</td>
<td>143 ± 23</td>
<td>141 ± 23</td>
<td>147 ± 23</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Peripheral diastolic BP (mmHg)</td>
<td>84 ± 11</td>
<td>82 ± 11</td>
<td>80 ± 11</td>
<td>78 ± 11</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Acute coronary syndrome (%)</td>
<td>31.6</td>
<td>29.6</td>
<td>29.6</td>
<td>34.5</td>
<td>0.202</td>
</tr>
<tr>
<td>Coronary artery disease (%)</td>
<td>10 % visual stenosis</td>
<td>78.7</td>
<td>81.4</td>
<td>83.6</td>
<td>86.2</td>
</tr>
<tr>
<td>Antihypertensive treatment (%)</td>
<td>86.1</td>
<td>83.1</td>
<td>84.0</td>
<td>94.0</td>
<td>0.001</td>
</tr>
<tr>
<td>ACE-inhibitors (%)</td>
<td>44.2</td>
<td>46.5</td>
<td>50.1</td>
<td>72.6</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>ARB&lt;sup&gt;e&lt;/sup&gt; (%)</td>
<td>3.3</td>
<td>3.7</td>
<td>4.1</td>
<td>6.9</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Beta blockers (%)</td>
<td>74.1</td>
<td>62.9</td>
<td>60.2</td>
<td>56.4</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Calcium-channel blockers (%)</td>
<td>15.5</td>
<td>17.0</td>
<td>13.9</td>
<td>16.3</td>
<td>0.876</td>
</tr>
<tr>
<td>Diuretics (%)</td>
<td>15.7</td>
<td>21.9</td>
<td>25.8</td>
<td>50.4</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>eGFR&lt;sup&gt;f&lt;/sup&gt; (mL/min per 1.73 m&lt;sup&gt;2&lt;/sup&gt;)</td>
<td>85.4 ± 22.3</td>
<td>88.6 ± 25.4</td>
<td>86.7 ± 24.4</td>
<td>80.9 ± 26.8</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>NYHA-classification (%)</td>
<td>1</td>
<td>56.2</td>
<td>53.7</td>
<td>53.2</td>
<td>44.4</td>
</tr>
<tr>
<td>2</td>
<td>28.0</td>
<td>30.1</td>
<td>30.2</td>
<td>28.4</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>13.1</td>
<td>14.2</td>
<td>13.9</td>
<td>22.6</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>2.7</td>
<td>2.0</td>
<td>2.7</td>
<td>4.6</td>
<td></td>
</tr>
<tr>
<td>LVEFg, %</td>
<td>63.1 ± 15.2</td>
<td>62.6 ± 16.2</td>
<td>60.2 ± 16.6</td>
<td>53.2 ± 19.3</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>NT-pBNP (ng/mL)</td>
<td>327.0 (138.0–678.5)</td>
<td>246.0 (101.5–686.0)</td>
<td>236.0 (83.0–699.0)</td>
<td>397.0 (114.0–1189.0)</td>
<td>0.769</td>
</tr>
<tr>
<td>25-hydroxyvitamin D (µg/L)</td>
<td>16.8 (11.2–23.9)</td>
<td>15.7 (10.0–22.9)</td>
<td>15.4 (10.3–22.6)</td>
<td>14.6 (9.0–22.0)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>High sensitivity CRP (mg/L)</td>
<td>2.7 (1.1–7.1)</td>
<td>2.9 (1.2–7.3)</td>
<td>3.1 (1.2–8.2)</td>
<td>4.9 (1.9–11.2)</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Values are given as median (25th, 75th percentile) for continuous variables, and percentage for categorical data.

Group differences (P for trend) were calculated by ANOVA for continuous and χ<sup>2</sup> test for categorical variables.

Quartiles are from baseline plasma renin concentration.

PRC, plasma renin concentration.

Ir-Angiotensin II, immunoreactive angiotensin II.

PAC, plasma aldosterone concentration.

Arterial hypertension was defined as systolic blood pressure >140 mmHg and/or diastolic blood pressure >90 mmHg and/or ongoing intake of antihypertensive medication.

ARB, angiotensin-II type-1 receptor blockers.

eGFR, estimated glomerular filtration rate according to the MDRD (modification of diet in renal disease) formula to estimate GFR.

LVEF, left ventricular ejection fraction.

Values are related to higher survival probability. Assumptions underlying the Cox proportional hazard models i.e. the proportionality of hazards were evaluated using log-minus-log survival and partial (Schoenfeld) residuals vs. survival time plots and were found valid. For cardiovascular mortality, the C statistic for the fully adjusted Model 3 was 0.692 (95% CI 0.666–0.717) without PRC and 0.693 (95% CI 0.668–0.718) with PRC, indicating that the addition of PRC to establish CVD risk factors does not significantly improve prediction of CVD mortality. Kaplan–Meier analysis showed an increased probability of cardiovascular death during follow-up
with increasing quartiles of PRC (Figure 1). Patients with high PRC levels continued to be separated throughout the follow-up period of almost 10 years (log-rank \( P \leq 0.001 \)).

In order to consider a varying relation of plasma renin to mortality with time additional fully adjusted Cox proportional hazard analyses with varying follow-up (below and above follow-up of 2.5 years) were performed. We found that the association between PRC expressed as a continuous variable and risk of fatal CVD events did not vary significantly at 2.5 years and after 2.5 years of follow-up; multivariable-adjusted HR per SD increment of log-PRC were at/below 2.5 years of follow-up 1.22 (95% CI 1.06–1.40, \( P = 0.005 \)) and >2.5 years of follow-up 1.20 (1.09–1.33, \( P < 0.001 \)).

Association of plasma renin with specific causes of cardiovascular disease and non-cardiovascular disease death

In fully adjusted Cox analyses for specific causes of CVD death, continuous log-PRC was associated with a 22% increase of risk of SCD per SD increment of log-PRC were at/below 2.5 years of follow-up 1.22 (95% CI 1.05–1.41, \( P = 0.009 \)) and >2.5 years of follow-up 1.22 (95% CI 1.06–1.40, \( P = 0.005 \)).

Plasma renin concentration associated cardiovascular disease mortality in various subgroups

To determine whether the association between PRC and CVD mortality was homogeneous across the cohort, we conducted subgroup analyses with formal testing for interactions.

We found a statistically significant interaction term between PRC-related CVD mortality and age, NT-pBNP levels, CAD, treatment with ACEi, beta-blockers, diuretics, and eGFR levels (Figure 2A/B). In brief, among those who were above the age of 60 years at baseline 1-SD increase of log-PRC was associated with a 26% higher risk of fatal CVD events, whereas no significant association between PRC and fatal CVD events was found for those younger than 60 years. Furthermore, participants revealing NT-pBNP levels

---

**Table 2** Cox proportional hazard ratios (95% CI) for cardiovascular events according to plasma renin concentration quartiles and according to per SD increment of log-plasma renin concentration increase

<table>
<thead>
<tr>
<th>Plasma renin concentration quartiles</th>
<th>Increment per log-SD PRC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Range (pg/mL)</td>
<td></td>
</tr>
<tr>
<td>1st quartile (&lt;5.5)</td>
<td>1.0 reference</td>
</tr>
<tr>
<td>2nd quartile (5.5–11.4)</td>
<td>1.15 (0.89–1.48) ( P = 0.276 )</td>
</tr>
<tr>
<td>3rd quartile (11.5–24.0)</td>
<td>1.26 (0.98–1.62) ( P = 0.077 )</td>
</tr>
<tr>
<td>4th quartile (&gt;24.0)</td>
<td>1.63 (1.28–2.07) ( P &lt; 0.001 )</td>
</tr>
</tbody>
</table>

**Cardiovascular mortality**

- **Crude model**: 1.0 reference 1.28 (0.99–1.64) \( P = 0.060 \)
- **Model 1**: 1.0 reference 1.26 (0.98–1.62) \( P = 0.077 \)
- **Model 2**: 1.0 reference 1.12 (0.85–1.48) \( P = 0.405 \)
- **Model 3**: 1.0 reference 1.49 (1.07–2.08) \( P = 0.019 \)

Model 1 adjusted for age and sex.
Model 2 additionally adjusted for systolic blood pressure, HDL- and LDL-cholesterol, ex and active smokers, coronary artery disease, body mass index, high-sensitivity C-reactive protein, NT-pBNP, 25-hydroxyvitamin D, diabetes mellitus type 1/2, the use of ACE-inhibitors, angiotensin-II type-1 receptor blockers, beta-blockers, calcium channel blockers, diuretics, and eGFR.
Model 3 additionally adjusted for immunoreactive angiotensin II and plasma aldosterone concentration.
>400 pg/mL at baseline were at 43% higher risk of CVD mortality per SD increase of log-PRC compared with those with NT-pBNP levels at/below 400 pg/mL. Moreover, 1 SD increase of log-PRC was related to a 22% higher risk of CVD mortality in those presenting with a CAD at baseline. In addition, a strong relation between higher risk of fatal CVD events and PRC was exclusively found in those showing an eGFR < 60 mL/min per 1.73 m² compared with those with eGFR levels at/higher than 60 mL/min per 1.73 m². Although we found a significant interaction for treatment with ACEi at baseline, PRC was related to higher risk of fatal CVD events in both subgroups with and without ACEi treatment, respectively. In participants with the use of beta-blockers and the use of diuretics, 1 SD increased of log-PRC was associated with a 32 and 29% higher risk of cardiovascular mortality, respectively. In contrast, no significant association was found in those without taking beta-blockers and diuretics, respectively. No effect modification was observed in case of severe HF (NYHA III/IV; \( P \) for interaction 0.060), LVEF (\( P \) for interaction 0.113), arterial hypertension (\( P \) for interaction 0.492), history of arterial hypertension (\( P \) for interaction 0.345), ACS (\( P \) for interaction 0.855), the use of calcium channel blockers (\( P \) for interaction 0.323), and the use of ARBs (\( P \) for interaction 0.426).

### Discussion

The main finding from our study is that baseline plasma renin levels were associated with higher short- and long-term CVD mortality independent of established CVD risk factors, antihypertensive medication, ir-Ang II, and plasma aldosterone levels. Analyses of specific causes of CVD death revealed strong associations between PRC and higher risk of SCD and death due to HF. The relationship between PRC and risk of fatal CVD events was modified by age, kidney function, NT-pBNP, the use of ACEi, the use of beta-blockers, the use of diuretics, and the presence of CAD.

The robust association between PRC and higher risk of CVD mortality independent of downstream components of the RAAS and ACE inhibition supports the emerging notion of vascular and myocardial damaging properties of renin. Activation of the ubiquitously expressed PRR by prorenin and renin may lead to a range of cellular events independent of Ang II and aldosterone. The (Pro)renin receptor activation in animals resulted in upregulation of pro-fibrotic- and pro-inflammatory genes such as transforming growth factor-β1, plasminogen activator inhibitor-1 (PAI-1), tumour necrosis factor (TNF)-α, fibronectin, and interleukin-1β, which are strongly involved in cardiac and vascular remodelling. This is in line with a strong association between plasma renin and higher risk of SCD and death due to HF.

Previous studies exploring the relation between plasma renin and CVD risk revealed conflicting results. Our findings suggest that age might potentially modify the association between plasma renin and incident CV death. Meade et al. did not see any associations between PRC and incident fatal CVD events by studying industrial workers with an age ranging from 40 to 60 years. A recently published investigation in CHF patients with a median age of 56 also failed to reveal an association between plasma renin and higher risk of mortality after consideration of various factors.
covariates. Our results therefore suggest that higher age might be an important prerequisite of renin-mediated CV damage. Whether the effect modification by ageing is due to an altered renin synthesis, dysregulated tissue RASs, higher prevalence of co-morbidities, or amplified activation of the PRR remains matter of research.

In contrast to previous investigations, we found that the risk of fatal CVD events in patients with higher PRC levels is independent of both arterial hypertension and positive history of arterial hypertension. This extends the finding of Alderman et al., who observed a strong association between plasma renin and higher risk of incident MI in hypertensive patients. In LURIC we could not identify a relation between PRC and higher risk of fatal MI. However, participants in LURIC were significantly older, BP was lower, and CAD was more frequently observed compared with the cohort of Alderman et al. Our findings suggest that a pre-existing vascular damage and impaired LV function might be important effect modifiers of PRC-associated CVD mortality. We documented that LV dysfunction reflected by higher NT-pBNP levels strongly interacts with renin-related fatal CV events. This is in line with decreased NT-pBNP levels after direct renin inhibition with aliskiren in addition to ACEi and beta-blockers in patients with CHF (NYHA II–IV).

We have recently documented increased plasma renin and aldosterone levels in patients with lower kidney function. In the present analysis, lower kidney function might be an important effect modifier of renin-related vascular and myocardial damage. The inappropriate activation of the RAS as well as insufficient inhibition of renin during ongoing treatment with ACEi and/or ARBs in the setting of chronic kidney disease might therefore result in a persistently high mortality within this patient group. Accordingly, in animal models with chronic kidney disease renin inhibition by aliskiren reduced mortality and end-organ damage. Our results support these findings by demonstrating that renin is related to CV death in low-eGFR patients independent of ir-Ang II and aldosterone. This further underlines potential renal, vascular, and myocardial protective effects of aliskiren in patients with kidney damage.

Renin–angiotensin–aldosterone system blockade by ACEi, ARBs, and mineralocorticoid receptor blockers have improved various clinical outcomes in a wide range of CHF patients. Residual deaths in patients with CAD, CHF, chronic kidney disease, and MI, however, remain high despite ongoing treatment with ACEi or ARBs. Pharmacological RAAS-blocking treatment strategies result in a reactive rise of renin due to impaired feedback inhibition by Ang II on renin formation. As a consequence, it has been suggested that inappropriately elevated renin levels might contribute to persistently higher rates of hospitalization, kidney damage, and CV deaths in various CV risk groups. Although we observed a significant interaction of PRC-related CVD mortality risk and the use of ACEi, we found an independent association between higher PRC and fatal CVD events in participants with and without intake of ACEi at baseline, respectively. This finding indicates that renin inhibition might exert tissue protective effects independent of an ongoing ACEi therapy, which has been confirmed in the ALOFT study. These and other findings further support the notion that renin-related CV risk might be independent of reactive risk due to less feedback inhibition during ACE inhibition. In line with this, Huang et al. documented that renin might potentially contribute to the development of vascular and myocardial damage such as increasing the formation of PAI-1 and TNF-β even in the absence of Ang II. In this context, our findings put renin into the perspective of an independent CVD risk factor and indicate beneficial effects of a renin-inhibiting treatment. Of note, the use of beta-blockers and the use of diuretics have been revealed as potential effect modifiers of PRC-related risk of fatal cardiovascular events. However, we want to stress that these subgroup and interaction analyses should be interpreted with caution. It should be considered that patient groups with a certain antihypertensive drug may not only differ to those without the respective drug intake by medication use per se but rather by co-morbidities that primarily indicated the drug prescription.

A row of currently ongoing randomized, placebo-controlled trials evaluate whether direct renin inhibition by aliskiren prevents the progression of CV and myocardial disease. Recently published findings from the ASPIRE trial may argue against additional beneficial effects of a dual RAS-blockade including aliskiren. Solomon et al. documented no significant change of functional and structural LV parameters between aliskiren and placebo treatment after 36 weeks in post-MI patients. Our findings support the suggestions by Solomon et al. that renin is associated with CV damage in the long term, indicating that longer treatment periods with renin inhibition might be necessary to exert organ protective effects.

**Limitations**

Our sample was primarily composed of elderly Caucasians of European ancestry referred to coronary angiography; thus, generalizations to other ethnicities and younger individuals cannot be made. The use of antihypertensive medication with varying doses might have impacted on the association between PRC levels and CVD mortality. Furthermore, despite extensive adjustments of the statistical models used to evaluate the association between PRC and CVD mortality, we cannot exclude residual confounding by co-morbidities such as HF or lower kidney function, which are implicated to impact on both PRC levels and cardiovascular outcome. Although, 24 h urine sodium excretion, which is an important mediator of renal renin release, was not determined, we attempted to consider various traditional and novel mediators of renin release in our analyses. Biomarkers used in statistical models were measured only once at baseline. Finally, the observational design of our study precludes conclusions with regard to causal relationships. Despite these limitations, our study has several strengths, particularly the in-depth clinical and biochemical characterization of the patients. Multiple established risk factors representing key pathological pathways implicated in the pathogenesis of CVD were available for conjoint and comparative multivariable analyses. Finally, our results are strengthened by the high number of participants eligible for mortality analyses and a considerable number of deaths within a long-term follow-up of almost 10 years.

Higher plasma renin levels are strongly and independently related to higher CVD risk, SCD, and death due to HF in patients referred to coronary angiography. The association between renin and incident CVD death remained stable independent of arterial hypertension, ACS, antihypertensive treatment, ir-Ang II, and
aldosterone. Higher NT-pBNP levels, pre-existing CAD, kidney function, age, and the use of renin-modulating antihypertensive drugs might be crucial effect modifiers for renin-mediated target organ damage. Further studies should therefore evaluate whether renin inhibition exerts organ protective effects and decreases mortality in various CV risk groups.

Acknowledgements

We thank the LURIC team involved in patient recruitment, sample and data handling, and the laboratory staffs at the Ludwigshafen General Hospital, the Universities of Freiburg, Ulm, and Graz.

Funding

LURIC has received funding from the 6th Framework Program (LURIC Study–a resource for functional genomics, grant LSHM-CT-2004-503485) and 7th of Framework Program (integrating project AtheroRemo, Grant Agreement number 201668) of the European Union.

Conflict of interest:

none declared.

References