Uric Acid Levels Are Associated With All-Cause and Cardiovascular Disease Mortality Independent of Systemic Inflammation in Men From the General Population

The MONICA/KORA Cohort Study

Christa Meisinger, Wolfgang Koenig, Jens Baumert, Angela Döring

Objective—The purpose of this study was to assess whether increasing serum uric acid (UA) levels are related to cardiovascular disease (CVD) mortality, all-cause mortality, and incident (fatal and nonfatal) myocardial infarction (MI) in men from the general population taking into account C-reactive protein (CRP), a sensitive marker of systemic inflammation.

Methods and Results—The study was based on 3604 men (45 to 74 years of age) who participated in 1 of the 3 MONICA Augsburg surveys between 1984 and 1995. All participants were prospectively followed within the framework of the Cooperative Health Research in the Region of Augsburg (KORA). Up to December 31, 2002, there occurred 809 total deaths, 359 CVD deaths, and 297 incident MIs. In a Cox model, comparing extreme quartiles of the UA distribution, the hazard ratio for CVD mortality was 1.44 (95% confidence interval [CI] 1.04 to 2.0), and for all-cause mortality it was 1.40 (95% CI 1.13 to 1.74) after adjustment for conventional cardiovascular risk factors, CRP, and diuretic intake. However, UA was not associated with incident MI after multivariable adjustment.

Conclusions—High UA levels were independently associated with CVD mortality as well as all-cause mortality but not with incident MI in middle-aged men from the general population. (Arterioscler Thromb Vasc Biol 2008;28:1186-1192)

Key Words: men ■ mortality ■ risk factors ■ serum uric acid ■ C-reactive protein

Several studies have investigated whether serum uric acid (UA) is an independent marker of cardiovascular disease (CVD) risk.1–8 Whereas most prospective studies found a positive association between serum UA and CVD mortality as well as all-cause mortality,1,2,7 in a few other studies UA levels were not associated with these outcomes.3,8 Because of these inconclusive findings, controversy remains about the nature of the relation between UA and cardiovascular events. Because of the large number of risk factors involved in CVD and their close relation to UA, it is difficult to prove or disprove an independent relationship. Differences in measurement of and adjustment for potential confounding factors in various studies could have contributed to the discrepancies. Thus, studies which extensively assess patients’ risk factor profiles are necessary to investigate whether UA plays a causal role in the development of CVD.

It is well known that serum UA is associated with the metabolic syndrome, which in turn is related to endothelial dysfunction, vascular inflammation, and hypertension—all of which may contribute to atherosclerosis.9,10 Very recently, a positive and significant association between UA and several inflammatory markers such as white blood cell count, C-reactive protein (CRP), interleukin (IL)-6, IL-18, and tumor necrosis factor (TNF)-α, was found in a large population-based sample of older persons.11 Furthermore, evidence suggests that UA might stimulate inflammation, which is clearly involved in the pathogenesis of CVD.12

We sought to investigate the predictive role of serum UA levels in a population-based sample of men, 45 to 74 years of age, with respect to long-term CVD mortality, all-cause mortality, and fatal as well as nonfatal incident myocardial infarction (MI) accounting for a large number of cardiovascular risk factors and the use of diuretics. Furthermore, we explored whether the relationship could be confounded by CRP, a sensitive systemic marker of inflammation which has been consistently associated with a variety of cardiovascular end points including myocardial infarction, stroke, and CVD mortality.13,14

Methods

Data was derived from the population-based MONICA (Monitoring trends and determinants on cardiovascular diseases) Augsburg
(Southern Germany) studies conducted between 1984 and 1995. The MONICA Augsburg project was part of the multinational WHO MONICA project, and the design of the project has been described in detail elsewhere. Three independent cross-sectional surveys were carried out in the city of Augsburg and the counties Augsburg and Aichach-Friedberg in 1984/85 (S1), 1989/90 (S2) , and 1994/95 (S3) to estimate the prevalence and distribution of cardiovascular risk factors among men and women. Altogether 13,427 men and women (response 77%) 25 to 74 years of age participated in at least 1 of the 3 cross-sectional studies. All persons who took part in more than 1 survey were included only once with data collected at the first visit. All subjects were prospectively followed within the framework of the Cooperative Health Research in the Region of Augsburg (KORA). The present study was restricted to 45- to 74-year-old male study participants (n = 3939 men), because CRP values were not measured in women and younger men. For the present analyses using mortality as outcome, we excluded all subjects with incomplete data on any of the included variables (n = 389). Thus, the prospective analyses comprised 3604 men 45 to 74 years of age at baseline for the outcomes all-cause and CVD mortality.

For the analyses using incident MI as outcome we excluded 354 study participants with incomplete data on any of the included variables and 215 persons with a prevalent MI at baseline. Thus, these analyses comprised 3424 men 45 to 74 years of age at baseline. Written informed consent was obtained from each study participant and the study was approved by the local ethics committee.

Data Collection
Baseline information on socio-demographic variables, smoking habits, leisure time physical activity level, medication use, and alcohol consumption were gathered by trained medical staff during a standardized interview. During the interview participants were also asked whether they suffer from diabetes, and if the diagnosis was made by a physician. In addition, all participants underwent an extensive standardized medical examination including the collection of a blood sample. All measurement procedures have been described elsewhere in detail. Body mass index (BMI) was calculated as weight in kilograms divided by height in square meters. Participants were classified as active during leisure time if they regularly participated in sports in summer and winter and if they were active for at least 1 hour per week in either season. Actual hypertension was defined as blood pressure values ≥ 140/90 mm Hg or use of antihypertensive medication given that subjects were aware of being hypertensive. Dyslipidemia was defined as the ratio of total cholesterol to high density cholesterol ≥ 5.0. The abbreviated Modification of Diet in Renal Disease Study Group equation was used to calculate estimated glomerular filtration rate (eGFR). History of CVD was defined as prevalent MI or prevalent stroke at baseline.

Clinical Chemical Measurements
A nonfasting venous blood sample was obtained from all study participants while sitting. Total serum cholesterol analyses were carried out using an enzymatic method (CHOD-PAP; Boehringer Mannheim). HDL cholesterol was also measured enzymatically after precipitation of the apoprotein B-containing lipoproteins with phosphotungstate/Mg2+/EDTA (Boehringer Mannheim). Serum creatinine was determined using an automated Jaffe method in S1 and S2 (Technicon, SMAC autoanalyzer) and an enzymatic method in S3 (creatinine PAP, Boehringer Mannheim). The enzymatic method was calibrated according to the Jaffe method. Serum UA was measured by the uricase method in S1 and S2. In S3, serum UA was determined with an enzymatic colorimetric reaction (Uric Acid PAP; Boehringer Mannheim). Samples for measurement of CRP were stored at −70°C until analysis. Serum CRP concentrations were measured with the use of an hs-immunoradiometric assay (range, 0.05 to 10 mg/L), as previously described.

Outcomes
The end points used in this study were incidence of fatal or nonfatal MI including sudden cardiac death, mortality from any CVD, and all-cause mortality. Deaths were ascertained by regularly checking the vital status of all sampled persons of the MONICA surveys through the population registries inside and outside the study area. Death certificates were obtained from local health departments and coded for the underlying cause of death by a single trained person using the 9th revision of the International Classification of Diseases (ICD-9). The outcomes used in this study were mortality from any CVD (ICD-9: 390 to 459) and all-cause mortality. MIs were identified through the population-based MONICA/KORA Augsburg coronary event registry which monitors the occurrence of all in- and out-of-hospital fatal and nonfatal MIs among the 25- to 74-year-old inhabitants of the study region.

Statistical Analyses
The association of ordinal UA quartiles and baseline characteristics were assessed by tests for trend using Mantel-Haenszel Chi² test for categorized characteristics and simple linear regression for continuous characteristics. Age-adjusted partial Pearson correlation coefficients (r) were calculated to assess associations between UA and continuous variables. The study population was stratified into 4 groups of UA concentrations with use of cut points of 4.9, 5.7, and 6.6 mg/dL (25th, 50th, and 75th percentiles). The distributions of CRP and creatinine concentrations were markedly skewed to the right and therefore log-transformed in analyses where normality was required. The proportional hazards assumption was valid for all factors used in the Cox models described below shown by parallel lines of log(log(event)) versus log of event times. Relative risks of incident MI, CVD, and all-cause mortality were computed for quartiles 2, 3, and 4, as compared with the lowest quartile in Cox proportional hazards models: the first model included UA and in addition age (continuous) and survey. The second model included all previous factors plus actual hypertension (yes/no), dyslipidemia (yes/no), level of leisure time physical activity (active/inactive), smoking status (regular smoking, that is a subject who smoked at least one cigarette per day at baseline, yes/no), alcohol intake (0, >0 and ≤ 40, or >40 g/d), BMI (continuous), history of diabetes (yes/no and unknown), history of CVD (for mortality outcomes only), serum creatinine (continuous, log-transformed), and diuretic use. The third model included in addition to all previous variables CRP (continuous, log-transformed). Interactions between UA and CRP as well as age were examined using likelihood ratio test which compared the -2 log (likelihood) between the model which contained only the main effects and the model which contained both the main effects and the interaction term. A test for trend was carried out by assigning median values to each quartile and including this variable in the Cox regression. To examine the joint effect of UA and CRP on CVD and all-cause mortality, combined UA and CRP variables were created. For this purpose, the upper quartile value of the UA measurement (6.6 mg/dL) was used as the cut point to define a low and high group. The cut point 3.0 mg/L was chosen for CRP to define a low and high group. Subjects were classified into 4 categories (low/low, low/high, high/low, and high/high). Those with low UA and low CRP values were chosen as the reference group.

Univariate survival analysis was performed with the use of cumulative event curves. Comparisons between survival curves were performed using Log Rank test. Results are presented as hazard ratios (HRs) and 95% confidence intervals (CIs). Significance tests were 2 tailed, and probability values less than 0.05 are stated as statistically significant. All analyses were performed using the Statistical Analysis System (Version 9.1, SAS Institute Inc).

Results
Between 1984 and 2002 (median follow-up period 11.7 years) 809 men died, 359 from CVD. Furthermore, there occurred 297 incident MIs during follow-up. The baseline characteristics of the study sample according to serum UA are shown in Table 1. Higher UA levels were associated with higher prevalences of obesity, and hypertension, more frequent regular smokers, and dyslipidemia. By contrast, men...
with higher UA were less frequently physically active during leisure time and less often reported diabetes. Higher UA was associated with higher alcohol intake, more advanced age, higher CRP values, and reduced eGFR. Furthermore, increased UA was related to more frequent use of diuretics.

When examined as continuous variables, serum UA was positively correlated with log-transformed CRP ($r=0.17$, $P<0.0001$), systolic blood pressure ($r=0.13$, $P<0.0001$), total cholesterol ($r=0.13$, $P<0.0001$), BMI ($r=0.22$, $P<0.0001$), and log-transformed serum creatinine ($r=0.12$, $P<0.0001$), and showed a negative age-adjusted correlation with HDL cholesterol ($r=-0.12$, $P<0.0001$).

As shown in Table 2 UA was significantly associated with CVD mortality and all-cause mortality. UA levels in the fourth quartile were significantly associated with CVD mortality (HR 1.44; 95% CI, 1.04 to 2.00) and all-cause mortality (HR 1.40; 95% CI, 1.13 to 1.74). There was no significant interaction between UA and CRP in this model.

Furthermore, a formal test for interaction between UA and age in the different models revealed no significant interaction.

In the age- and survey-adjusted analysis (Model 1), UA was also significantly associated with incident MI (HR 1.48; 95% CI, 1.06 to 2.05) for the highest versus lowest quartile of UA. However, after multivariable adjustment the initially significant association between UA and incident MI was strongly attenuated and no longer statistically significant (Model 3: HR 1.10; 95% CI 0.77 to 1.57; Table 2).

Cumulative event probabilities for CVD mortality and all-cause mortality are presented in Figure 1, with $P<0.0001$ from log-rank tests of significance across quartiles of serum UA.

The joint relationships between UA and CRP values are shown in Figure 2. The combination of both high UA and high CRP was associated with a statistically significantly increased risk for CVD mortality (HR 1.74; 95% CI 1.26 to 2.40) and for all cause mortality (HR 1.73; 95% CI 1.39 to 2.16) compared with both parameters not being high (referent) in the multivariable adjusted model.

Discussion

In this large prospective population-based cohort study, middle-aged men with high UA levels showed an unfavorable

### Table 1. Means (SD) and Prevalences of Baseline Variables According to Quartiles of Uric Acid, 45- to 74-Year-Old Men

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>01: &lt;4.9</th>
<th>02: 4.9 to &lt;5.7</th>
<th>03: 5.7 to &lt;6.6</th>
<th>04: ≥6.6</th>
<th>P Value for Trend</th>
</tr>
</thead>
<tbody>
<tr>
<td>Men, n=3604</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age, y</td>
<td>57.3 (8.0)</td>
<td>58.0 (7.9)</td>
<td>57.6 (8.3)</td>
<td>58.5 (8.2)</td>
<td>0.0080</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>26.7 (3.2)</td>
<td>27.5 (3.1)</td>
<td>28.3 (3.5)</td>
<td>28.8 (3.8)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Systolic blood pressure, mm Hg</td>
<td>135.4 (17.3)</td>
<td>138.3 (18.9)</td>
<td>139.5 (18.0)</td>
<td>142.3 (19.7)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Diastolic blood pressure, mm Hg</td>
<td>81.2 (10.4)</td>
<td>83.7 (11.5)</td>
<td>83.6 (11.0)</td>
<td>84.7 (12.3)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Actual hypertension, %</td>
<td>46.5</td>
<td>52.5</td>
<td>57.0</td>
<td>65.5</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Serum creatinine, mg/dL*</td>
<td>0.95 (1.2)</td>
<td>1.05 (1.2)</td>
<td>0.98 (1.2)</td>
<td>1.00 (1.3)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>eGFR, ml/min/1.73 m²</td>
<td>89.9 (23.0)</td>
<td>88.8 (21.0)</td>
<td>86.8 (22.9)</td>
<td>84.1 (22.6)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>CRP, mg/L*</td>
<td>1.32 (3.3)</td>
<td>1.44 (3.0)</td>
<td>1.83 (2.8)</td>
<td>2.19 (2.8)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Total cholesterol, mg/dL</td>
<td>236.2 (42.2)</td>
<td>242.1 (41.9)</td>
<td>245.1 (44.2)</td>
<td>251.0 (50.6)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>HDL cholesterol, mg/dL</td>
<td>52.6 (15.5)</td>
<td>51.0 (13.8)</td>
<td>47.8 (14.4)</td>
<td>48.4 (16.5)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>TC/HDL ratio ≥5.0, %</td>
<td>36.8</td>
<td>46.0</td>
<td>55.1</td>
<td>57.5</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Regular smoking, %</td>
<td>21.0</td>
<td>20.4</td>
<td>27.3</td>
<td>25.5</td>
<td>0.0014</td>
</tr>
<tr>
<td>Alcohol intake 0 g/d, %</td>
<td>21.8</td>
<td>18.7</td>
<td>16.6</td>
<td>12.1</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>≥0.1–0.99 g/d, %</td>
<td>56.2</td>
<td>51.9</td>
<td>49.3</td>
<td>43.0</td>
<td></td>
</tr>
<tr>
<td>≥40 g/d, %</td>
<td>22.0</td>
<td>29.5</td>
<td>34.1</td>
<td>44.9</td>
<td></td>
</tr>
<tr>
<td>Physically active, %</td>
<td>39.7</td>
<td>39.7</td>
<td>36.7</td>
<td>32.8</td>
<td>0.0008</td>
</tr>
<tr>
<td>History of diabetes, %</td>
<td>10.9</td>
<td>7.0</td>
<td>6.6</td>
<td>4.6</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Education (&lt;12 years), %</td>
<td>71.1</td>
<td>67.0</td>
<td>69.7</td>
<td>73.0</td>
<td>0.2104</td>
</tr>
<tr>
<td>Use of diuretics, %</td>
<td>7.2</td>
<td>6.6</td>
<td>10.7</td>
<td>17.6</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

*Geometric mean.
CVD risk profile. However, the prevalence of diabetes decreased with increasing UA levels. This is in accordance with prior studies demonstrating that UA concentrations in diabetic patients are significantly lower than in nondiabetic subjects, most likely attributable to an increased renal uric acid clearance in diabetic persons; however, the underlying mechanisms have not been fully elucidated. Serum UA levels were strongly associated with CVD mortality and all-cause mortality but not with incident MI after adjustment for a variety of known cardiovascular risk factors. Further adjustment for CRP did not appreciably attenuate the associations which remained statistically significant. Despite the positive correlation between UA and CRP, in joint analyses, the highest risk for both CVD and all-cause mortality was observed in men with elevated levels of both biomarkers. The findings of the present study contribute important new epidemiological evidence to earlier findings on associations of serum UA with CVD death and death from any cause among middle-aged initially healthy men by taking into account CRP as a measure of systemic inflammation.

Association Between UA and CVD in Earlier Prospective Studies

Several earlier studies have suggested that serum UA is associated with increased CVD and all-cause mortality. In the NHANES I study for example increased serum UA was strongly associated with the risk of CVD mortality independent of cardiovascular risk factors, alcohol consumption, and diuretic intake. Furthermore, Niskanen et al reported a 2.5-fold increased risk of CHD mortality in the upper versus the lower serum UA tertile among 1423 healthy middle-aged Finnish men. Also, an earlier report from the MONICA/KORA cohort showed that baseline serum UA levels were an independent predictor of CVD mortality and all-cause mortality in 45- to 64-year-old men. However, in several other studies UA was not associated with these outcomes. Because in some of them the association between UA and CVD disappeared after multivariable adjustment, it has been suggested that UA may not play a role in the etiology of CVD. Consequently, the idea that hyperuricemia may be a secondary phenomenon continued to be reasonable. Although not significant in the present study it seems that men with low and high levels of UA had higher risk of mortality than those with average UA levels. Because of the relatively small sample size this finding could be by chance. However, a J-shaped relation between UA and mortality was also found in other studies, for example by Verdecchia et al in subjects with hypertension, by Lehto et al in subjects with type 2 diabetes, and by Culleton et al in men from the general population. In the present study there was no significant relationship between UA levels and incident fatal and nonfatal MI. This finding confirms previous studies and is also in agreement with a very recent investigation, which could show that UA is associated with certain subforms of CVD only.

Potential Proatherogenic Effects of UA

Recent insights into the biological effects of UA have refuted the idea that it is a biologically inert molecule. It has been demonstrated that UA is linked to insulin resistance, that it...
may enhance oxidation of low-density lipoprotein cholesterol, and lipid peroxidation,\textsuperscript{23} and may increase platelet adhesiveness.\textsuperscript{24} Mercuro et al found an intrinsic negative effect of UA on the arterial wall in hyperuremic subjects.\textsuperscript{25} Moreover, UA has been identified as a mediator of endothelial dysfunction.\textsuperscript{26} On the other hand, studies have proposed strong antioxidant properties of UA.\textsuperscript{27} Because atherosclerosis has been linked to increased oxidative stress, hyperuricemia might represent a compensatory mechanism to protect the body from prooxidants.\textsuperscript{28} However, in the metabolic syndrome environment, UA becomes a strong oxidant,\textsuperscript{29} a phenomenon recently named as "urate redox shuttle."\textsuperscript{29,30}

Very recently, it could be shown that UA plays a role in immune activation\textsuperscript{31} and cytokine secretion.\textsuperscript{32} Furthermore, Ruggiero et al reported that UA was positively and significantly associated with several inflammatory markers in a
large population-based sample of persons aged 65 to 95 years. Because the association between UA and proinflammatory markers was found also in subjects with normal UA levels it could be assumed that UA is not only a marker of catabolic rate but might also be actively involved in systemic inflammation, which plays an important role in the development of atherothrombosis. In addition, it could be shown that baseline UA and changes in UA during 3 years of follow-up predicted changes in circulating CRP independent of relevant confounders.

In the present study, including 45- to 74-year-old men, the association between UA and mortality was independent of CRP. This finding is contrary to an earlier study conducted in a sample of community-dwelling older people 70 to 79 years of age, which also took into account the potential confounding effect of CRP and IL-6. In that study, no independent association between serum UA level and all-cause mortality in older men and women was found. Also, the authors concluded that underlying inflammation as measured by serum levels of CRP and IL-6 may be an important covariate in this context. In our study, no significant interaction was found between UA level and age, thus it is unlikely that the relationship between UA and mortality differs by age groups.

The strengths of the MONICA/KORA Augsburg Cohort Study are primarily its prospective design, the representativeness of the cohort, based on a large random sample of the general population and the availability of extensive data on lifestyle and multiple cardiovascular risk factors for which we carefully adjusted.

Conclusion
The present study provides further evidence for an independent association between elevated UA and risk of CVD death as well as death from any cause in apparently healthy middle-aged men from the general population. However, further studies are needed to investigate the pathophysiological mechanisms by which high serum UA is associated with atherosclerotic vascular disease. The measurement of UA, an easily available and inexpensive risk marker, might turn out as a valuable tool for improved individual cardiovascular risk assessment, in addition to conventional risk factors.

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Disclosures

None.

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