# Leptin, adiponectin, their ratio and risk of coronary heart disease: results from the MONICA/KORA Augsburg study 1984–2002

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# Leptin, adiponectin, their ratio and risk of coronary heart disease: Results from the MONICA/KORA Augsburg Study 1984–2002

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## 1. Introduction

Leptin and adiponectin are obesity-related hormones and cytokines, so-called adipokines, produced by adipocytes. Despite modulating a number of metabolic processes linked to atherosclerosis [1], including glucose regulation [2], insulin sensitivity [3], hematopoiesis [4], fatty acid catabolism [5] and angiogenesis [6], their potential association with coronary heart disease (CHD) is still a matter of controversy [7,8].

Leptin is a pleiotropic adipokine with structural and functional relation to proinflammatory cytokines and is widely expressed, including monocytes, myocardial cells [9], and atherosclerotic lesions [10]. It induces proinflammatory cytokines like MCP-1 [5], elicits macrophage foam cell formation [11], and promotes platelet aggregation and arterial thrombosis in obesity [12].

Adiponectin has antiinflammatory effects on the cellular components of the vascular wall by inhibiting NF-kappaB signaling [13]. It protects against myocardial ischemia-reperfusion injury through AMPK- and COX-2-dependent mechanisms, and induces nitric oxide expression from endothelial cells [14]. Furthermore, exogenous adiponectin administration protects ApoE-deficient mice from atherosclerosis [15].

Although results from the aforementioned experimental studies seem to be fairly consistent, data of epidemiological studies remain somewhat controversial [8,16]. While various studies investigating the association between circulating adipokine levels and CHD yielded fairly strong associations, several other investigations were not able to demonstrate any meaningful relationship with CHD [17,18].

We sought to further elucidate the role of leptin and adiponectin in the prediction of incident CHD (fatal and nonfatal myocar-

*Abbreviations:* CHD, coronary heart disease; CRP, C-reactive protein; FU, follow-up; HR, hazard ratio; HDL-C, HDL-cholesterol; IL-6, interleukin-6; IRMA, immunoradiometric assay; KORA, Kooperative Gesundheitsforschung in der Region Augsburg (Cooperative Health Research in the Region of Augsburg); MI, myocardial infarction; MONICA, monitoring of trends and determinants in cardiovascular diseases; RR, relative risk; S1, MONICA/KORA baseline survey 1 conducted in 1984–1985; S2, MONICA/KORA baseline survey 2 conducted in 1989–1990; S3, MONICA/KORA baseline survey 3 conducted in 1994–1995; SCD, sudden cardiac death; sE-selectin, soluble E-selectin; sICAM-1, soluble intercellular adhesion molecule-1; TC, total cholesterol.

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**Table 1**
Baseline demographic, clinical, and laboratory characteristics of the participants with and without incident CHD during follow-up.

| Characteristic | Men | | | Women | | |
|---|---|---|---|---|---|---|
| | CHD Cases | Noncases | $p^*$ | CHD Cases | Noncases | $p^*$ |
| Number | 253 | 827 | | 80 | 901 | |
| Age | 57.0 (0.53) | 52.1 (0.39) | <.001 | 58.2 (0.75) | 52.5 (0.35) | <.001 |
| Education (<12 years) % | 77.1 (0.03) | 66.9 (0.02) | <.001 | 86.5 (0.04) | 85.1 (0.01) | 0.741 |
| Smoking status (%) | | | <.001 | | | 0.378 |
| Current smoker | 43.1 (0.03) | 29.1 (0.02) | | 24.3 (0.05) | 18.4 (0.01) | |
| Former smoker | 38.1 (0.03) | 41.5 (0.02) | | 18.4 (0.04) | 16.6 (0.01) | |
| Never smoker | 18.8 (0.02) | 29.4 (0.02) | | 57.3 (0.06) | 65.1 (0.02) | |
| Frequency of exercise (%) | | | <.001 | | | <.001 |
| Active | 30.4 (0.03) | 42.5 (0.02) | | 18.8 (0.04) | 35.7 (0.02) | |
| Inactive | 69.6 (0.03) | 57.5 (0.02) | | 81.2 (0.04) | 64.3 (0.02) | |
| Alcohol consumption† ( %) | | | 0.175 | | | 0.038 |
| 0g/d | 20.9 (0.03) | 17.6 (0.01) | | 59.1 (0.06) | 44.0 (0.02) | |
| <39.9/19.9 g/d | 43.4 (0.03) | 50.1 (0.02) | | 27.5 (0.05) | 35.7 (0.02) | |
| ≥40/20 g/d | 35.7 (0.03) | 32.2 (0.02) | | 13.4 (0.04) | 20.3 (0.01) | |
| Body mass index (kg/m$^2$) | 28.1 (0.24) | 27.3 (0.13) | 0.004 | 29.5 (0.58) | 26.8 (0.15) | <.001 |
| Waist-to-hip ratio‡ | 0.95 (<.01) | 0.93 (<.01) | <.001 | 0.84 (0.01) | 0.81 (<.01) | <.001 |
| Parental history of MI (%) | | | 0.018 | | | 0.415 |
| Positive | 24.0 (0.03) | 18.1 (0.01) | | 20.6 (0.05) | 21.6 (0.01) | |
| Unknown | 26.6 (0.03) | 22.0 (0.01) | | 27.2 (0.05) | 20.2 (0.01) | |
| Negative | 49.5 (0.03) | 59.9 (0.02) | | 52.2 (0.06) | 58.2 (0.02) | |
| History of actual hypertension (%) | 62.6 (0.03) | 44.2 (0.02) | <.001 | 73.5 (0.05) | 38.5(0.02) | <.001 |
| Systolic blood pressure (mmHg) | 141.7 (1.26) | 135.8 (0.64) | <.001 | 147.2 (2.58) | 131.7 (0.68) | <.001 |
| Diastolic blood pressure (mmHg) | 83.6 (0.74) | 83.4 (0.38) | 0.823 | 83.9 (1.62) | 79.9 (0.37) | 0.014 |
| Current HRT§ | | 5.5 (0.03) | 10.4 (0.01) | 0.109 | | |
| Ratio TC/HDL | 5.83 (0.13) | 5.07 (0.06) | <.001 | 5.40 (0.29) | 4.02 (0.04) | <.001 |
| C-reactive protein (mg/L)‖ | 2.46 (1.07) | 1.44 (1.04) | <.001 | 2.79 (1.13) | 1.44 (1.04) | <.001 |
| Interleukin-6 (pg/mL)‖ | 3.08 (1.06) | 2.12 (1.04) | <.001 | 3.40 (1.10) | 1.90 (1.04) | <.001 |
| sICAM-1, ng/mL | 869,7 (22.3) | 785.3 (10.8) | 0.001 | 879.8 (36.7) | 729.8 (8.6) | <.001 |
| sE-selectin (ng/mL) | 66.10 (2.51) | 59.78 (1.04) | 0.020 | 65.05 (3.35) | 51.22 (0.80) | <.001 |
| Adiponectin (μg/mL)‖ | 9.47 (1.02) | 9.44 (1.01) | 0.905 | 12.51 (1.04) | 13.41 (1.01) | 0.112 |
| Leptin (ng/mL)‖ | 5.53 (1.05) | 4.86 (1.03) | 0.024 | 22.44 (1.09) | 16.73 (1.03) | 0.001 |

Data are weighted percentages for categorical variables, weighted means (standard errors) for normally distributed continuous variables and ‖weighted geometric means with (antilog of standard errors of log means) for skewed continuous variables.

* The *t*-test for continuous variables and $x^2$ test for categorical variables.

† Men: 0, >0 to 39.9 g/d, ≥40 g/d; Women: 0, >0 to 19.9 g/d, ≥20 g/d.

§ Only for women aged ≥50 years ($n$ = 585) with no current use of OC.

‡ Only measured in participants of survey 2 and 3 (cases:$n$ = 229; noncases:$n$ = 1170).

Weights:cases = all cases/non-missing cases; noncases = 1/sampling fraction with sampling fraction = subcohort/full cohort without cases for each sex and survey.

HRT, hormone replacement therapy.

dial infarction (MI) and sudden cardiac death (SCD)) in a large prospective population-based cohort study of middle-aged men and women. The present study investigates the association of leptin and adiponectin levels and incident CHD in the general population. Furthermore, unlike previous studies, we also sought to analyze the combined effect of leptin and adiponectin using the leptin/adiponectin ratio, since it has been suggested to provide a better indicator of atherosclerosis risk than each adipokine alone [19].

## 2. Methods

### 2.1. Study population

The monitoring of trends and determinants in cardiovascular disease (MONICA)/Cooperative Health Research in the Region of Augsburg (KORA) studies served as the database for a prospective case-cohort study in initially healthy, middle-aged men and women [20]. Briefly, three independent population-based MONICA/KORA Augsburg surveys (S), with a total number of 13,427 participants (6725 men, 6702 women) aged 25–64 (S1) or 25–74 years (S2–S3), were conducted in 1984/85 (S1), 1989/90 (S2) and 1994/95 (S3). All subjects were prospectively followed within the framework of KORA. The case-cohort design used in the present study has been described previously in detail [21].

Due to the low incidence of CHD under the age of 35, the present study was limited to 10,718 persons (5382 men and 5336 women) between 35 and 74 years of age at baseline who participated in at least one of the three surveys. After exclusion of 1187 subjects with missing blood samples and 231 participants with self-reported, prevalent CHD, the source population for the present study comprised 9300 subjects (4507 men, 4793 women).

For the case-cohort study, a random sample of the source population, called here the subcohort, containing 2163 subjects (1154 men, 1009 women) was selected stratifying by sex and survey. Participants with missing values for leptin, adiponectin or any of the covariables used in the present analysis were excluded leading to a subcohort of 1820 subjects (901 men, 919 women). The final stratum-specific sample sizes were used together with the stratum-specific sizes of the cohort of interest to compute sampling fractions, and the inverse of the sampling fractions yielded the survey and sex specific sampling weights: 4.63, 4.28, 6.56 for men, and 4.41, 5.06, 6.45 for women. A variant of these weights was used for calculations of weighted means and proportions based on the cohort random sample and additional incident CHD cases.

### 2.2. Assessment of risk factors for cardiovascular disease, ascertainment of CHD at follow-up, statistical methods

All assessment procedures and standard laboratory methods have been described elsewhere [22]. Serum levels of interleukin-6 (IL-6), soluble E-Selectin (sE-selectin) and soluble intercellular adhesion molecule-1 (sICAM-1) were determined as previously described using commercially available ELISAs [23,24].

**Table 2**
Weighted Pearson correlation coefficients between leptin, adiponectin, the leptin/adiponectin ratio and selected biomarkers for CHD, based on the randomly cohort sampled subcohort, adjusted for sex.

| Characteristics | Log leptin | | Log adiponectin | | Log leptin/adiponectin ratio | |
|---|---|---|---|---|---|---|
| | *R* | *p* | *R* | *p* | *R* | *p* |
| Log leptin | – | – | −0.157 | <0.001 | 0.932 | <0.001 |
| Log adiponectin | −0.157 | <0.001 | – | – | −0.504 | <0.001 |
| Log Il-6 | 0.200 | <0.001 | −0.079 | <0.001 | 0.204 | <0.001 |
| Log CRP | 0.361 | <0.001 | −0.139 | <0.001 | 0.367 | <0.001 |
| Age | 0.183 | <0.001 | 0.232 | <0.001 | 0.075 | 0.001 |
| BMI | 0.661 | <0.001 | −0.171 | <0.001 | 0.640 | <0.001 |
| sE-selectin | 0.162 | <0.001 | −0.070 | 0.003 | 0.167 | <0.001 |
| sICAM-1 | 0.065 | 0.006 | 0.013 | 0.589 | 0.052 | 0.026 |
| Systolic blood pressure | 0.245 | <0.001 | −0.003 | 0.892 | 0.215 | <0.001 |
| Diastolic blood pressure | 0.220 | <0.001 | −0.078 | <0.001 | 0.221 | <0.001 |
| Physical activity | −0.093 | <0.001 | −0.035 | 0.132 | −0.069 | 0.003 |
| TC/HDL-C ratio | 0.249 | <0.001 | −0.284 | <0.001 | 0.322 | <0.001 |
| Waist-to-hip ratio* | 0.389 | <0.001 | −0.164 | <0.001 | 0.400 | <0.001 |

* $n = 1228$.

C-reactive protein (CRP) concentrations were measured with a high-sensitivity immunoradiometric assay (IRMA) [25] or a high-sensitivity latex-enhanced nephelometric assay on a BN II analyzer (Dade Behring, Marburg, Germany) as previously described in more detail [26]. Serum levels of leptin and adiponectin were both measured by ELISAs from Mercodia, Uppsala, Sweden. The intra- and inter-assay coefficients of variation were <10.0%. All analyses were run in a blinded fashion.

For all analyses a $p$-value <0.05 was considered to be statistically significant. All statistical evaluations were performed using the SAS software package (Version 9.1, SAS-Institute, Cary, NC, USA).

Most aspects of data collection and laboratory measurements have been described before. For additional details and information on statistical analyses, please see the online data supplement (www.sciencedirect.com).

## 3. Results

Overall, 2061 participants (333 subjects with incident CHD and 1728 subjects without incident CHD) of the 3 population-based MONICA/KORA Augsburg surveys were included in this case – cohort study. The mean FU time (±SD) was 10.8 (±4.6) years. The baseline demographic, clinical, and laboratory characteristics of the study population are shown in Table 1. Subjects with incident CHD were older, were less likely to be never smokers, were less active, and showed a higher body mass index (BMI) and waist-to-hip ratio (WHR) compared with noncases. Furthermore, cases more frequently reported hypertension, and less likely negative parental history of MI, whereas significant differences for educational levels only were observed in men and significant differences in alcohol consumption between the two groups only in women. As expected, total cholesterol (TC)/high-density lipoprotein cholesterol (HDL-C) ratio was considerably higher in cases compared with noncases. Furthermore, concentrations of C-reactive protein (CRP) and interleukin-6 (IL-6) were higher in CHD cases than in noncases; ($p < 0.001$ for both parameters). Both, leptin and adiponectin levels were higher in women. Geometric means of leptin were 5.5 (1.1) ng/L in male and 22.4 (1.1) ng/L in female cases and 4.9 (1.0) in male and 16.7 (1.0) in female noncases, and for adiponectin in cases they were 9.5 (1.0) μg/mL for men and 12.5 (1.0) μg/mL for women and in noncases 9.4 (1.0) μg/mL for men and 13.4 (1.0) μg/mL for women.

Pearson correlation coefficients ($R$) adjusted for sex were calculated between log leptin and inflammatory markers, lipid variables or conventional risk factors based on the randomly sampled subcohort of 1820 individuals. They revealed a positive, statistically significant, correlation between log leptin and log IL-6, log CRP, ICAM-1 and E-selectin (Table 2). Furthermore, a positive correlation between log leptin and age, body mass index (BMI), TC/HDL-C ratio and waist-to-hip ratio and systolic and diastolic blood pressure and a negative correlation with log adiponectin and physical activity were also observed. Strongest correlations were found for BMI ($R = 0.66$), waist-to-hip ratio ($R = 0.39$) and log CRP ($R = 0.36$).

Adjusted Pearson correlation coefficients ($R$) between log adiponectin and inflammatory markers, lipid variables or conventional risk factors revealed an inverse, statistically significant, correlation between log adiponectin and log CRP. Furthermore significant inverse correlations between log adiponectin and BMI, TC/HDL-C ratio, waist-to-hip ratio, IL-6, E-selectin, diastolic blood pressure and a positive correlation with age were also observed. None of these correlation coefficients reached an absolute value above 0.3.

Correlations of the ratio of leptin/adiponectin with risk factors were similar to those observed for leptin.

Table 3 shows the results of Cox proportional hazards analysis, in which the association of baseline levels of leptin with incident CHD was assessed. In age, sex and survey adjusted analyses (Model 1), there was a statistically significant association between increased concentrations of leptin and incident CHD (HR regarding top vs bottom tertile, 1.35; 95% CI, 1.00 to 1.83; $P$ for trend 0.013), whereas this association was no longer statistically significant after further adjustment for BMI, smoking status, physical activity and alcohol intake (Model 2) (HR, 1.07; 95% CI, 0.74 to 1.53; $P$ for trend 0.484). The association was even further attenuated after additional adjustment for systolic blood pressure, TC/HDL-C ratio and parental history (Model 3) (HR, 0.88; 95% CI, 0.60 to 1.29; $P$ for trend 0.758). Adjustment for inflammatory markers and markers of endothelial

**Table 3**
Hazard ratios (95% CI) for the risk of incident CHD according to baseline concentrations of leptin.

| Tertiles of leptin | | | | | | |
|---|---|---|---|---|---|---|
| | T1 | T2 | | T3 | | p for trend |
| | HR | HR | 95% CI | HR | 95% CI | |
| Model 1* | 1.0 | 1.03 | 0.75–1.41 | 1.35 | 1.00–1.83 | 0.013 |
| Model 2† | 1.0 | 0.91 | 0.65–1.27 | 1.07 | 0.74–1.53 | 0.484 |
| Model 3‡ | 1.0 | 0.78 | 0.55–1.11 | 0.88 | 0.60–1.29 | 0.758 |
| Model 4§ | 1.0 | 0.76 | 0.54–1.09 | 0.79 | 0.53–1.17 | 0.887 |

* Adjustment for age, sex, survey.
† Additional adjustment for BMI, smoking status, physical activity, alcohol consumption.
‡ Additional adjustment for systolic BP, TC/HDL-C ratio, parental history of MI.
§ Additional adjustment for CRP, IL-6, ICAM-1, sE-selectin.

**Table 4**
Hazard ratios (95% CI) for the risk of incident CHD according to baseline concentrations of adiponectin.

| Tertiles of Adiponectin | | | | | | |
|---|---|---|---|---|---|---|
| | T1 | T2 | | T3 | | p for trend |
| | HR | HR | 95% CI | HR | 95% CI | |
| Model 1* | 1.0 | 0.94 | 0.69–1.27 | 1.42 | 1.06–1.91 | 0.025 |
| Model 2† | 1.0 | 0.88 | 0.64–1.21 | 1.19 | 0.86–1.64 | 0.318 |
| Model 3‡ | 1.0 | 0.77 | 0.55–1.06 | 0.91 | 0.65–1.28 | 0.632 |
| Model 4§ | 1.0 | 0.75 | 0.54–1.05 | 0.87 | 0.62–1.23 | 0.492 |

* Adjustment for age, sex, survey.
† Additional adjustment for BMI, smoking status, physical activity, alcohol consumption.
‡ Additional adjustment for systolic BP, TC/HDL-C ratio, parental history of MI.
§ Additional adjustment for CRP, IL-6, ICAM-1, sE-selectin.

**Table 5**
Hazard ratios (95% CI) for the risk of incident CHD according to baseline leptin/adiponectin ratio.

| Tertiles of leptin/adiponectin ratio | | | | | | |
|---|---|---|---|---|---|---|
| | T1 | T2 | | T3 | | p for trend |
| | HR | HR | 95% CI | HR | 95% CI | |
| Model 1* | 1.0 | 1.39 | 1.01–1.91 | 1.78 | 1.30–2.43 | <0.001 |
| Model 2† | 1.0 | 1.26 | 0.88–1.78 | 1.44 | 1.00–2.09 | 0.051 |
| Model 3‡ | 1.0 | 1.13 | 0.78–1.61 | 1.13 | 0.77–1.66 | 0.352 |
| Model 4§ | 1.0 | 1.06 | 0.73–1.54 | 1.01 | 0.68–1.51 | 0.650 |

* Adjustment for age, sex, survey.
† Additional adjustment for BMI, smoking status, physical activity, alcohol consumption.
‡ Additional adjustment for systolic BP, TC/HDL-C ratio, parental history of MI.
§ Additional adjustment for CRP, IL-6, ICAM-1, sE-selectin.

dysfunction (Model 4) further attenuated the relation between elevated leptin concentrations and risk of subsequent coronary events. No interactions between tertiles of leptin or adiponectin and sex were indicated using likelihood ratio tests. Results for the sex specific analyses are shown in the online supplement Table 1.

Conversely to leptin, as Table 4 shows, low levels of adiponectin were associated with incidence of CHD after adjustment for age, sex and survey (Model 1). However, after additional adjustment for coronary risk factors (models 2–4), the association was no longer significant. Again, there was no significant sex interaction. Stratified analyses are shown in the online supplement Table 2.

Table 5 shows, that the leptin/adiponectin ratio was also significantly associated with the risk of incidence CHD in model 1 (HR, 1.78; 95% CI, 1.30 to 2.43; *P* for trend <0.001), whereas further adjustment for BMI, smoking status, physical activity and alcohol intake (Model 2) decreased the HR and it became borderline non-significant (HR, 1.44; 95% CI, 1.00 to 2.09; *P* for trend 0.051). In the final model 4, the HR comparing top versus bottom tertiles of the leptin/adiponectin ratio was non-significant and no sex specific differences were noted (Online supplement Table 3).

Furthermore we analyzed the hazard ratios (95% CI) for incident CHD according to baseline concentrations of leptin and adiponectin stratified by BMI (Table 6). For leptin there was no significant interaction with BMI, while elevated adiponectin concentrations seemed to be protective in overweight, but not in normal weight and obese, subjects.

## 4. Discussion

The present study reports the potential association between leptin and adiponectin and incident CHD in initially healthy middle-aged men and women from the general population. Our findings indicate no association between leptin, adiponectin and their ratio with the risk of CHD after adjustment for potential confounders.

### 4.1. Leptin and risk of CHD

In a prospective case-control study of hypertensive men and women, serum leptin was compared in 171 patients with MI and in 342 matched controls. Baseline leptin concentrations were significantly higher among patients with MI compared to matched controls (25.1 versus 20.0 ng/mL, $P = 0.007$). This difference remained significant after adjustment for traditional cardiovascular risk factors [27]. Welsh et al. measured leptin levels in a cohort of 5672 patients with vascular disease and could not show an association with cardiovascular disease risk in multivariable adjusted analyses in men or women [28].

These results however cannot be directly compared to our findings, since they were based on subjects with pre-existing end-stage renal disease or vascular disease, whereas our data came from mainly healthy subjects drawn from the general population.

Studies in patients without pre-existing vascular disease also yielded discrepant results. While a nested case-control study from WOSCOPS (West of Scotland Coronary Prevention Study), in 377 male cases and 783 controls, reported a 20% increase in CHD per one standard deviation (SD) increase in leptin levels after multivariable adjustment [29], Sattar et al. only found a moderate and statistically non-significant association between leptin levels and CHD risk that was further attenuated following adjustment for BMI [17]. Concordantly with this finding, recently it was shown in a moderate-sized community-based elderly sample that higher circulating leptin levels were associated with a greater risk of cardiovascular disease, but leptin did not provide incremental prognostic information beyond BMI [30]. In a meta-analysis of available prospective studies on circulating leptin levels and CHD risk, involving a total of 1335 cases and 3407 controls, the combined risk ratio comparing extreme thirds of leptin levels across all studies was 2.28 (95% CI, 1.42 to

**Table 6**
Hazard ratios (95% CI) for incident CHD according to baseline concentrations of leptin and adiponectin stratified by BMI.

| Tertiles of leptin | | | | | | | |
|---|---|---|---|---|---|---|---|
| | T1 | T2 | | T3 | | p trend | p interaction |
| | HR | HR | 95% CI | HR | 95% CI | | |
| Normal weight (BMI <25 kg/m$^2$) | 1.0 | 0.60 | 0.28–1.26 | 0.84 | 0.22–3.21 | 0.389 | 0.471 |
| Overweight (BMI 25–30 kg/m$^2$) | 1.0 | 0.85 | 0.21–1.43 | 0.68 | 0.39–1.18 | 0.922 | |
| Obese (BMI ≥30 kg/m$^2$) | 1.0 | 2.18 | 0.33–14.50 | 2.88 | 0.43–19.40 | 0.635 | |
| Tertiles of adiponectin | | | | | | | |
| Normal weight (BMI <25 kg/m$^2$) | 1.0 | 0.80 | 0.36–1.78 | 1.96 | 0.83–4.62 | 0.189 | 0.012 |
| Overweight (BMI 25–30 kg/m$^2$) | 1.0 | 0.80 | 0.49–1.31 | 0.52 | 0.29–0.95 | 0.032 | |
| Obese (BMI ≥ 30 kg/m$^2$) | 1.0 | 0.80 | 0.39–1.64 | 1.16 | 0.58–2.31 | 0.569 | |

Adjustment for age, survey, sex, physical activity, alcohol consumption, systolic BP, TC/HDL-C ratio, parental history of MI, CRP, IL-6, sICAM-1, sE-selectin.

3.68) in analyses adjusted for sex and age only, and was attenuated to 1.44 (95% CI, 0.95 to 2.16) after further adjustment for potential confounders [17].

### 4.2. Adiponectin and risk of CHD

An inverse association of adiponectin with coronary heart disease (CHD) has been reported from several studies. However, meta-analysis of seven prospective studies involving a total of 1318 CHD cases, only yielded an odds ratio of 0.84 (95% CI, 0.70 to 1.01) comparing extreme tertiles of adiponectin [18]. Consistent with previous data we report that circulating adiponectin levels are inversely associated with waist-to-hip ratio, BMI, the TC/HDL cholesterol-ratio, the inflammatory markers CRP and Il-6 and the endothelial marker sE-selectin. However, in contrast to various previous reports, and despite the observed moderate, but significant, inverse correlations between adiponectin levels and BMI/waist-to-hip ratio, the present study, involving 2061 subjects, indicates that low levels of circulating adiponectin do not predict future risk of CHD after adjustment for potential confounders. Nethertheless, elevated adiponectin concentrations seemed to be protective in overweight.

### 4.3. Leptin/adiponectin ratio and risk of CHD

To the best of our knowledge no previous prospective study examined the leptin/adiponectin ratio with regard to CHD risk. Pearson correlation coefficients show strikingly similar results between leptin and the leptin/adiponectin ratio. In this study, after fully adjustment, the ratio does not yield any additional information on CHD risk compared to leptin. Our data is in contrast to a cross-sectional study in Italian subjects, which suggested that the leptin/adiponectin ratio might be a better marker of intima media thickness of the carotid artery than either adiponectin or leptin alone [19]. However, it should be pointed out that cross-sectional observations are prone to several biases, therefore further prospective data are needed to confirm this observation.

### 4.4. Limitations and strengths of the study

This study has several limitations that need to be addressed. First, the mean FU period of 11 years is relatively long and might be responsible for the weakening of the association between the risk marker and the disease outcome. Second, concentrations of leptin, adiponectin and their ratio were significantly correlated with the majority of other measured inflammatory markers and risk factors for CHD. Thus, it may be difficult to keep the single effects of one marker apart due to the collinearity. However, regression diagnostics revealed that there were no collinearity problems in the final models.

Our study has also several strengths which include the population-based prospective design conducted in initially healthy subjects, the large number of incident cases, the simultaneous measurement of several adipokines and inflammatory factors, a long follow-up period, the minimization of the likelihood of survival bias because fatal and nonfatal coronary events were included in our study, and the careful adjustment for conventional risk factors by multivariable methods.

## 5. Conclusion

In conclusion, adipokines are predictors of coronary events, but after adjustment for other coronary risk factors, we cannot demonstrate a statistically significant independent association between increased leptin and decreased adiponectin concentrations and subsequent coronary events in apparently healthy, middle-aged subjects in this large prospective case-cohort study. Moreover, our findings demonstrate that the ratio of these pro- and anti-inflammatory adipokines does not significantly improve prediction of incident CHD compared to either adipokine alone. Thus, leptin, adiponectin and the leptin/adiponectin ratio are unlikely to present major independent risk factors for CHD. However, the lack of prognostic impact of increased leptin and decreased adiponectin concentrations on the incidence of CHD among apparently healthy subjects does not completely exclude the potential significance of adipokine-regulated mechanisms in the pathophysiology of atherothrombotic disease, especially taking into account the potent plaque stabilizing and destabilizing properties of these molecules.

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## Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.atherosclerosis.2009.08.020.

## References

[1] Sommer G, Kralisch S, Stangl V, et al. Secretory products from human adipocytes stimulate proinflammatory cytokine secretion from human endothelial cells. J Cell Biochem 2009;106(4):729–37.
[2] Kamohara S, Burcelin R, Halaas JL, Friedman JM, Charron MJ. Acute stimulation of glucose metabolism in mice by leptin treatment. Nature 1997:374–7.
[3] Rossetti L, Massillon D, Barzilai N, et al. Short term effects of leptin on hepatic gluconeogenesis and in vivo insulin action. J Biol Chem 1997;272(44):27758–63.
[4] Lam QL, Lu L. Role of leptin in immunity. Cell Mol Immunol 2007;4:1–13.
[5] Yamagishi SI, Edelstein E, Du XL, et al. Leptin induces mitochondrial superoxide production and monocyte chemoattractant protein-1 expression in aortic endothelial cells by increasing fatty acid oxidation via protein kinase A. J Biol Chem 2001;276:25096–100.
[6] Anagnostoulis S, Karayiannakis AJ, Lambropoulou M, et al. Human leptin induces angiogenesis in vivo. Cytokine 2008;42(3):353–7.
[7] Martin SS, Qasim A, Reilly MP. Leptin resistance: a possible interface of inflammation and metabolism in obesity-related cardiovascular disease. J Am Coll Cardiol 2008;52:1201–10.
[8] Rathmann W, Herder C. Adiponectin and cardiovascular mortality: evidence for "reverse epidemiology". Horm Metab Res 2007;39:1–2.
[9] Margetic S, Gazzola C, Pegg GG, Hill RA. Leptin: a review of its peripheral actions and interactions. Int J Obes Relat Metab Disord 2002;26:1407–33.
[10] Kang SM, Kwon HM, Hong BK, et al. Expression of leptin receptor (Ob-R) in human atherosclerotic lesions: potential role in intimal neovascularization. Yonsei Med J 2000;41(1):68–75.

[11] OïRourke L, Gronning LM, Yeaman SJ, Shepherd PR. Glucose-dependent regulation of cholesterol ester metabolism in macrophages by insulin and leptin. J Biol Chem 2002;277, 42557-12562.
[12] Konstantinides S, Schafer K, Koschnik S, Loskutoff DJ. Leptin-dependent platelet aggregation and arterial thrombosis suggests a mechanism for atherothrombotic disease in obesity. J Clin Invest 2001;108:1533–40.
[13] Ouchi N, Kihara S, Arita Y, et al. Adiponectin, an adipocyte-derived plasma protein, inhibits endothelial NF-kappaB signaling through a cAMP-dependent pathway. Circulation 2000;102:1296–301.
[14] Shibata R, Sato K, Pimentel DR, et al. Adiponectin protects against myocardial ischemia-reperfusion injury through AMPK- and COX-2–dependent mechanisms. Nat Med 2005;11:1096–103.
[15] Yamauchi T, Kamon J, Waki H, et al. Globular adiponectin protected ob/ob mice from diabetes and ApoE-deficient mice from atherosclerosis. J Biol Chem 2003;278:2461–8.
[16] Koenig W, Khuseyinova N, Baumert N, Meisinger C, Löwel H. Serum concentrations of adiponectin and risk of type 2 diabetes mellitus and coronary heart disease in apparently healthy middle-aged men: results from the 18-year follow-up of a large cohort from southern Germany. J Am Coll Cardiol 2006;48:1369–77.
[17] Sattar N, Wannamethee G, Sarwar N, et al. Leptin and coronary heart disease. J Am Coll Cardiol 2009;53:167–75.
[18] Sattar N, Wannamethee G, Sarwar N, et al. Adiponectin and coronary heart disease: a prospective study and meta-analysis. Circulation 2006;114:623–9.
[19] Norata GD, Raselli S, Grigore L, et al. Leptin:adiponectin ratio is an independent predictor of intima media thickness of the common carotid artery. Stroke 2007;38(10):2844–6.
[20] Hense HW, Filipiak B, Döring A, et al. Ten-year trends of cardiovascular risk factors in the MONICA Augsburg Region in Southern Germany. Results from the 1984/85, 1989/90 and 1994/1995 surveys. CVD Prevention 1998;1:318–1327.
[21] Koenig W, Khuseyinova N, Baumert J, et al. Increased concentrations of C-reactive protein and Il-6 but not Il-18 are independently associated with incident coronary events in middle-aged men and women: results from the MONICA/KORA Augsburg case-cohort study, 1984-2002. Arterioscler Thromb Vasc Biol 2006;26(12):2745–51.
[22] Meisinger C, Thorand B, Schneider A, et al. Sex differences in risk factors for incident type 2 diabetes mellitus: the MONICA Augsburg cohort study. Arch Intern Med 2002;162(1):82–9.
[23] Müller S, Martin S, Koenig W, et al. Impaired glucose tolerance is associated with increased serum concentrations of interleukin 6 and co-regulated acute-phase proteins but not TNF-alpha or its receptors. Diabetologia 2002;45:805–12.
[24] Thorand B, Baumert J, Chambless L, et al. Elevated markers of endothelial dysfunction predict type 2 diabetes mellitus in middle-aged men and women from the general population. Arterioscler Thromb Vasc Biol 2006;26:398–405.
[25] Hutchinson WL, Koenig W, Fröhlich M, et al. Immunoradiometric assay of circulating C-reactive protein: age-related values in the adult general population. Clin Chem 2000;46:934–8.
[26] Thorand B, Baumert J, Kolb H, et al. Sex differences in the prediction of type 2 diabetes by inflammatory markers: results from the MONICA/KORA Augsburg case-cohort study, 1984–2002. Diabetes Care 2007;30:854–60.
[27] Wallerstedt SM, Eriksson AL, Niklason A, Ohlsson C, Hedner T. Serum leptin and myocardial infarction in hypertension. Blood Press 2004;13(4):243–6.
[28] Welsh P, Murray HM, Buckley BM, et al. Leptin predicts diabetes but not CVD: results from a large prospective study in the elderly. Diabetes Care 2009;32(2):308–10.
[29] Wallace AM, McMahon AD, Packard CJ, et al. Plasma leptin and the risk of cardiovascular disease in the west of Scotland coronary prevention study (WOSCOPS). Circulation 2001;104(25):3052–6.
[30] Lieb W, Sullivan LM, Roubenoff R, et al. Plasma leptin levels and incidence of heart failure, cardiovascular disease, and total mortality in elderly individuals. Diabetes Care 2009;32(4):612–6.