Sex Differences in the Prediction of Type 2 Diabetes by Inflammatory Markers

Results from the MONICA/KORA Augsburg case-cohort study, 1984–2002

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OBJECTIVE — Although sex differences have been reported for associations between obesity and inflammation, the question of whether there is an effect modification by sex in the association between inflammation and type 2 diabetes has not been investigated in detail. Therefore, the aim of this study was to compare associations of markers of inflammation with type 2 diabetes risk between men and women.

RESEARCH DESIGN AND METHODS — Following a case-cohort design, cases of incident type 2 diabetes were identified from 7,936 subjects aged 35–74 years at baseline who participated in the population-based Monitoring of Trends and Determinants in Cardiovascular Disease (MONICA)/Cooperative Research in the Region of Augsburg (KORA) studies conducted between 1984 and 2002. Concentrations of C-reactive protein (CRP) and interleukin (IL)-6 were measured in 527 cases of incident type 2 diabetes (305 men and 222 women) and 1,698 noncases (889 men and 809 women).

RESULTS — After adjustment for age and survey and lifestyle factors including smoking, alcohol intake, and physical activity, elevated concentrations of CRP showed a considerably stronger association with risk of type 2 diabetes in women (hazard ratio comparing tertile extremes 7.60 [95% CI 4.43–13.04]) than in men (1.84 [1.27–2.67]). The *P* value for the sex interaction was <0.001. Further adjustment for metabolic risk factors considerably attenuated these associations, and they became nonsignificant in men but remained significant in women. IL-6 was also more strongly associated with type 2 diabetes in women, but there was no significant sex interaction.

CONCLUSIONS — Our data suggest that inflammatory processes may be of particular importance in the pathogenesis of type 2 diabetes in women.

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Abbreviations: CRP, C-reactive protein; IL, interleukin; IRMA, immunoradiometric assay; KORA, Cooperative Research in the Region of Augsburg; MONICA, Monitoring of Trends and Determinants in Cardiovascular Disease; SHBG, sex hormone–binding globulin; WHR, waist-to-hip ratio.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

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Chronic subclinical inflammation has been suggested to be involved in the pathogenesis of type 2 diabetes (1,2). This hypothesis has recently been supported by several prospective studies showing that subjects who developed type 2 diabetes during the follow-up period had elevated levels of markers of inflammation such as C-reactive protein (CRP) or interleukin (IL)-6 at baseline compared with subjects who did not develop the disease (3–16). However, although various studies have examined these associations, few reported sex-specific results (5,11,15,16). This lack of data is surprising, since there is strong evidence for sex differences in associations between diabetes, obesity, endogenous sex hormones, and inflammation (17–21). Most of the studies that investigated sex differences concerning inflammatory markers and risk for type 2 diabetes were relatively small and yielded contradictory results, especially for CRP. While the Hoorn Study, conducted in the Netherlands, reported a significant association between CRP and incident diabetes in men but not in women (11), the opposite was seen in the Mexico City Diabetes Study (5). Finally, in two studies conducted in subjects of Japanese origin, CRP was significantly associated with incident type 2 diabetes in both sexes (15,16).

To further elucidate possible sex differences in the association between markers of inflammation and type 2 diabetes, we separately analyzed the association between CRP and IL-6 as markers of inflammation and incident type 2 diabetes in men and women from a large population-based study.

RESEARCH DESIGN AND METHODS

— The present study is based on data collected within the population-based Monitoring of Trends and Determinants in Cardiovascular Disease (MONICA)/Cooperative Research in the Region of Augsburg (KORA) studies. As part of the MONICA Augsburg project, three independent, cross-sectional, population-based surveys covering the city

area of Augsburg, Germany, and two adjacent counties were conducted between 1984 and 1995 to estimate the prevalence and distribution of cardiovascular risk factors among men and women aged 25–64 (first survey) or 25–74 (second and third survey) years. The study was approved by the local authorities, and all participants provided written informed consent. The total number of participants was 13,427 (6,725 men and 6,702 women). All subjects were prospectively followed within the frame of KORA. The present study was restricted to subjects aged 35–74 years at baseline, since the incidence of type 2 diabetes is low in younger subjects. Altogether, 10,718 individuals (5,382 men and 5,336 women) in this age range participated in at least one of the three baseline surveys. After exclusions, which have been described in detail elsewhere (22) and which included 14 subjects with incident diabetes other than type 2 diabetes, the source population for the present study comprised 7,936 subjects (3,894 men and 4,042 women). Of women in the source population, 55.8% were aged ≥50 years and can thus be considered postmenopausal.

Within the MONICA/KORA studies, we designed a prospective case-cohort study. We chose a case-cohort design as preferable to a nested case-control design because of our interest in more than one outcome, namely diabetes and coronary heart disease. The case-cohort design has been well described (23) and widely used (12,22,24,25) and includes a random sample of the cohort of interest (source population) and the incident cases for the disease of study, which is type 2 diabetes in this study and coronary heart disease in others (25).

In the present study, the stratified random sample of the source population, referred to here as the subcohort, contained 1,885 subjects (1,018 men and 867 women) and was selected separately for women and men, stratifying by survey. From these, we excluded 55 men and 12 women with missing values on CRP, IL-6, or any of the covariables used in the analyses, leading to a subcohort of 1,818 subjects (963 men and 855 women). Detailed information on the sample sizes by sex and survey and the calculation of the sex- and survey-specific sampling weights and methods used to identify self-reported incident cases of type 2 diabetes have been described elsewhere (22). Briefly, at follow-up, all subjects were asked whether they had diabetes and whether the disease had been diagnosed by a physician. In addition, the year of diagnosis was assessed. Cases with self-reported incident diabetes were validated by a questionnaire mailed to the treating physician or by medical chart review. Self-reported cases that could not be validated were excluded. The mean ± SD follow-up time was 10.8 ± 5.1 years.

A total of 555 validated incident cases of type 2 diabetes (329 men and 226 women) were observed until 31 December 2002. Of these, 24 men and 4 women were excluded because of incomplete information, leaving 305 men and 222 women with incident type 2 diabetes for the final analyses. Since 74 male and 46 female cases were also part of the randomly drawn subcohort, the present analysis comprised a total of 2,225 participants (305 men and 222 women with incident type 2 diabetes and 889 male and 809 female noncases).

Data collection and laboratory measurements

Standardized interviews were conducted by trained medical staff to assess information concerning sociodemographic variables, smoking habits, leisure time physical activity, and alcohol consumption. In addition, participants underwent standardized medical examinations including collection of a nonfasting venous blood sample. All assessment procedures and standard laboratory procedures have been described elsewhere (26–28). Blood samples stored at −80°C were used to analyze CRP and IL-6. CRP concentrations were measured using a high-sensitivity immunoradiometric assay (IRMA) (men aged 45–64 years) (29) or a high-sensitivity latex enhanced nephelometric assay on a BN II analyzer (men aged 35–44 years and all women) (Dade-Behring, Marburg, Germany). Both methods gave similar results when the same samples were analyzed. In a sample of 312 patients with clinically stable coronary artery disease and 476 voluntary blood donors, CRP concentrations ranged from 0.055 to 95 mg/l as measured by the IRMA and from 0.16 to 89 mg/l as measured by the Dade-Behring assay. The geometric mean CRP concentration was 1.31 mg/l for the IRMA and 1.17 mg/l for the Dade-Behring assay. The mean difference between the IRMA and the Dade-Behring assay was −0.39 mg/l with 95% limits of agreement of −1.88 and 1.11 mg/l when values >10 mg/l were excluded. The Deming regression coefficient between both methods was 0.94 (range 0–10 mg/l) (30). Serum levels of IL-6 were determined using a previously described sandwich enzyme-linked immunosorbent assay (31). The intra- and interassay coefficients of variation of quality control test sera for CRP and IL-6 were as follows: *1*) CRP-IRMA, 4.0 and 12.0%; *2*) CRP nephelometric assay, 2.5 and 5.1%; and 3) IL-6, <10.0 and <10.0%, respectively.

Statistical analysis

Cox proportional hazards analysis was used to assess the association between CRP and IL-6 and incident type 2 diabetes. Due to the case-cohort design, SEs were corrected using a SAS macro (SAS Institute, Cary, NC) with a sampling weight approach developed by Barlow (23). Sex-specific weighted tertiles of CRP and IL-6 in the subcohort were used to classify subjects in different risk groups. Results are presented for each tertile of CRP and IL-6, respectively (coded as dummy variables with the bottom tertile as the reference category), as hazard ratios (HRs) together with 95% CIs. *P* values are based on robust variance estimates using the Barlow macro. For testing for trends, tertiles were coded with their median values. Interactions between tertiles of CRP and IL-6 (coded as dummy variables) and sex were examined using likelihood ratio tests, which compared the −2 log (likelihood) between the model containing only the main effects and the model containing both the main effects and interaction terms. For all statistical analyses, $P < 0.05$ was considered to be statistically significant. All evaluations were performed with the statistical software package SAS version 8.02 for Unix and version 9.1 for Windows (for the procedure SURVEYFREQ).

RESULTS — Baseline characteristics of cases and noncases have been previously reported for a slightly different sample size (32) and are shown in Table 1 of the online appendix (available at http://dx.doi.org/10.2337/dc06-1693). Elevated concentrations of CRP measured at baseline were strongly associated with an increased risk of type 2 diabetes in both men and women in the age-, survey-, and lifestyle (i.e., smoking status, alcohol intake and physical activity)-adjusted analyses, but associations were considerably stronger in women (Table 1, model 2). HRs for the top tertile of CRP were 1.84 (95% CI 1.27–2.67) for men and 7.60

Table 1—*HRs for the risk of developing type 2 diabetes according to baseline levels of CRP for men and women*

	Men				Women			
	Tertile 1	Tertile 2	Tertile 3	*P* for trend	Tertile 1	Tertile 2	Tertile 3	*P* for trend
Median (range) (mg/l)	0.49 (0.05–0.84)	1.43 (0.85–2.31)	4.42 (2.32–80.21)		0.44 (0.10–0.82)	1.37 (0.83–2.37)	4.76 (2.39–47.00)	
Cases/noncases	60/313	106/286	139/290		17/287	65/273	140/249	
Model 1*	1.0	1.67 (1.17–2.40)	2.10 (1.48–3.00)	<0.001	1.0	3.37 (1.93–5.91)	7.90 (4.64–13.45)	<0.001
P value		0.005	<0.001			<0.001	<0.001	
Model 2†	1.0	1.60 (1.11–2.31)	1.84 (1.27–2.67)	0.007	1.0	3.44 (1.96–6.04)	7.60 (4.43–13.04)	<0.001
P value		0.01	0.001			<0.001	<0.001	
Model 3‡	1.0	1.26 (0.86–1.84)	1.13 (0.75–1.69)	0.97	1.0	2.55 (1.42–4.58)	3.91 (2.16–7.08)	<0.001
P value		0.24	0.55			0.002	<0.001	
Model 4§	1.0	1.21 (0.82–1.79)	1.09 (0.71–1.66)	0.94	1.0	2.06 (1.12–3.80)	2.74 (1.47–5.11)	0.01
P value		0.34	0.70			0.02	0.002	
Model 5	1.0	1.21 (0.82–1.80)	1.09 (0.71–1.67)	0.95	1.0	2.03 (1.10–3.75)	2.57 (1.36–4.84)	0.03
P value		0.33	0.69			0.02	0.004	

Data are HR (95% CI) unless otherwise indicated. Tertiles of the weighted distributions in the subcohort, stratified by sex, were used. *Adjusted for age and survey. †Additionally adjusted for lifestyle factors, i.e., smoking status (never smoker, former smoker, or current smoker), alcohol consumption (0, 0.1–39.9, or ≥40 g/day for men; 0, 0.1–19.9, or ≥20 g/day for women), and physical activity (inactive or active). ‡Additionally adjusted for BMI. §Additionally adjusted for systolic blood pressure, total–to–HDL cholesterol ratio, and parental history of diabetes (positive, unknown, or negative). ¶Additionally adjusted for IL-6. Models contained continuous variables unless otherwise indicated.

(4.43–13.04) for women. The *P* value for the interaction between tertiles of CRP and sex was <0.001. Further adjustment for BMI considerably lowered the observed HRs, and associations became nonsignificant in men but remained highly significant in women (model 3). Additional adjustment for systolic blood pressure, the ratio of total to HDL cholesterol, and parental history of type 2 diabetes (model 4) further attenuated the observed HRs, but associations remained highly significant in women (top tertile 2.74 [1.47–5.11]). The *P* value for the sex interaction was borderline significant in model 4 ($P = 0.06$). The addition of BMI as a squared term to the models did not lead to significant changes in the observed HRs (data not shown).

Elevated concentrations of IL-6 were also associated with incident type 2 diabetes in both men and women (Table 2). In age-, survey-, and lifestyle-adjusted analyses, men with IL-6 levels in the top tertile had a 2.29-fold increased risk of type 2 diabetes (95% CI 1.61–3.25) compared with men with IL-6 levels in the bottom tertile. The respective HR for women was 4.15 (2.61–6.62). Further adjustment for BMI and the other risk factors mentioned above (models 3 and 4) attenuated the HRs, but associations remained significant in men and women (HR for the top tertile in model 4: 1.58 [1.09–2.30] for men and 2.08 [1.23–3.51] for women). Sex differences were not statistically significant for any of these models.

Associations between both CRP and IL-6 and type 2 diabetes did not significantly change when model 4 was additionally adjusted for use of antihypertensive or lipid-lowering medication (online appendix Tables 2 and 3).

In women, adjustment for use of oral contraceptives and hormone replacement therapy in addition to the risk factors included in model 4 had almost no impact on the associations between CRP or IL-6 and incident type 2 diabetes (online appendix Tables 2 and 3). There was also no indication of a significant interaction between hormone replacement therapy use and CRP or IL-6 in the prediction of type 2 diabetes; however, one has to keep in mind that the number of women using hormone replacement therapy at baseline was relatively small.

When CRP and IL-6 were simultaneously included in the multivariable-adjusted Cox proportional hazards models, associations between CRP or IL-6

Table 2—*HRs for the risk of developing type 2 diabetes according to baseline levels of IL-6 for men and women*

	Men				Women				
	Tertile 1	Tertile 2	Tertile 3	*P* for trend	Tertile 1	Tertile 2	Tertile 3	*P* for trend	*P* for sex interaction
Median (range) (pg/ml)	1.11 (0.12–1.73)	2.33 (1.74–3.15)	4.85 (3.17–337.20)		0.94 (0.12–1.58)	2.19 (1.58–2.92)	4.39 (2.93–171.80)		
Cases/noncases	61/310	95/295	149/284		31/283	52/273	139/253		
Model 1*	1.0	1.64 (1.15–2.34)	2.58 (1.83–3.64)	<0.001	1.0	1.65 (1.03–2.66)	4.08 (2.62–6.36)	<0.001	0.12
P value		0.006	<0.001			0.04	<0.001		
Model 2†	1.0	1.56 (1.09–2.23)	2.29 (1.61–3.25)	<0.001	1.0	1.70 (1.05–2.75)	4.15 (2.61–6.62)	<0.001	0.08
P value		0.01	<0.001			0.03	< 0.001		
Model 3‡	1.0	1.31 (0.91–1.90)	1.68 (1.17–2.42)	0.008	1.0	1.42 (0.86–2.35)	2.52 (1.52–4.19)	<0.001	0.71
P value		0.15	0.005			0.17	<0.001		
Model 4§	1.0	1.22 (0.83–1.80)	1.58 (1.09–2.30)	0.02	1.0	1.39 (0.81–2.37)	2.08 (1.23–3.51)	0.004	0.79
P value		0.31	0.02			0.23	0.006		
Model 5¶	1.0	1.24 (0.84–1.83)	1.68 (1.14–2.46)	0.009	1.0	1.38 (0.81–2.37)	2.05 (1.19–3.53)	0.009	0.79
P value		0.28	0.008			0.24	0.01		

Data are HR (95% CI) unless otherwise indicated. Tertiles of the weighted distributions in the subcohort, stratified by sex, were used. *Adjusted for age and survey. †Additionally adjusted for lifestyle factors, i.e., smoking status (never smoker, former smoker, or current smoker), alcohol consumption (0, 0.1–39.9, or ≥40 g/day for men; 0, 0.1–19.9, or ≥20 g/day for women), and physical activity (inactive or active). ‡Additionally adjusted for BMI. §Additionally adjusted for systolic blood pressure, total–to–HDL cholesterol ratio, and parental history of diabetes (positive, unknown, or negative). ¶Additionally adjusted for CRP. Models contained continuous variables unless otherwise indicated.

and type 2 diabetes did not change appreciably (Tables 1 and 2, model 5).

In stratified analyses, we observed that associations between CRP, as well as IL-6, and type 2 diabetes tended to be stronger in men with a BMI <30 kg/m^2 (model 4, BMI included as a categorical variable with two categories: *P* value for interaction between CRP and BMI = 0.014; *P* value for interaction between IL-6 and BMI = 0.18). These differences were not apparent in women (*P* value for interaction between CRP and BMI = 1.00; *P* value for interaction between IL-6 and BMI = 0.67) (online appendix Table 4).

Stratification by smoking status revealed that, in women, the association between CRP and IL-6, respectively, and type 2 diabetes was stronger in ex-smokers and nonsmokers compared with current smokers. However, there was no significant interaction between smoking status and CRP or IL-6. In men, such a difference was not apparent. CRP and IL-6, respectively, were neither significantly associated with type 2 diabetes in smokers nor in ex- and nonsmokers (online appendix Table 4). In ex- and nonsmokers combined, *P* values for sex differences were similar to those obtained in the total study sample (*P* values were <0.001, <0.001, 0.02, 0.07, and 0.08 for CRP and 0.12, 0.16, 0.71, 0.55, and 0.54 for IL-6 for models 1, 2, 3, 4, and 5, respectively). There was no indication of a sex difference in current smokers for either CRP or IL-6 in the multivariable-adjusted models.

To determine the impact of abdominal body fat in addition to total body fat on the observed associations, we performed additional analyses in the subset of participants for whom measurements of waist-to-hip ratio (WHR) and waist circumference were available (men: 205 cases, 537 noncases; women: 128 cases, 491 noncases). Associations between inflammatory markers and incident type 2 diabetes were generally similar in this group compared with the total study sample, and adjustment for WHR or waist circumference in addition to BMI and other risk factors for type 2 diabetes had virtually no effect on the observed HRs. The only exception was the association between CRP and incident type 2 diabetes in women, in whom HRs were generally somewhat higher, and the addition of waist circumference led to attenuated HRs. The HR for the upper tertile of CRP compared with the lower tertile was 5.32

(95% CI 1.76–16.08) in the subgroup when only BMI and the other covariables of model 4 were included and decreased to 4.69 (1.55–14.18) when waist circumference was added. The additional inclusion of a squared term for WHR or waist circumference had almost no impact on the observed HRs (data not shown).

We conducted several sensitivity analyses. First, we excluded all subjects with ≤5 years of follow-up (men: n = 232, 121 cases, 111 noncases; women: n = 151, 76 cases, 75 noncases) to ensure that the observed associations were not caused by undiagnosed prevalent cases of diabetes at baseline. Multivariable-adjusted HRs estimated from these models were slightly lower than HRs estimated in the whole study sample (except for associations between IL-6 and type 2 diabetes in women, where slightly higher associations were observed), but associations remained highly significant for CRP and IL-6 in women (online appendix Tables 2 and 3). When follow-up time was limited to 5 years, HRs for associations between CRP and IL-6 were somewhat higher (except for IL-6 in women) but had wider CIs because of the smaller sample sizes (online appendix Tables 2 and 3). Second, we excluded all subjects with prevalent myocardial infarction, prevalent stroke, prevalent angina pectoris, or incident myocardial infarction (men: n = 182, 68 cases, 114 noncases; women: n = 83, 27 cases, 56 noncases) to determine any potentially confounding effect by the presence of cardiovascular disease at baseline. Again, the observed HRs were relatively similar in this group compared with the whole study sample except for the association between IL-6 and type 2 diabetes in men, which was somewhat attenuated and became nonsignificant (online appendix Tables 2 and 3).

CONCLUSIONS — In this population-based study, we demonstrated that CRP and IL-6 are associated with type 2 diabetes in both men and women. However, especially for CRP, associations were considerably stronger in women than in men. While the association between CRP and incident type 2 diabetes was strong and independent of other known risk factors for diabetes in middle-aged women, CRP was not independently associated with type 2 diabetes in men, as previously reported for fewer incident cases from the same study population (8). In contrast, IL-6 independently predicted the disease in both sexes.

Previous studies led to conflicting results as to whether there exists a sex difference in the association between a proinflammatory state and diabetes risk (5,11,15,16). Our observation that CRP is more strongly associated with type 2 diabetes in women than in men is in line with results from the Mexico City Diabetes Study (5). This notion is also supported by a reanalysis of data from the Insulin Resistance Atherosclerosis Study (33) and by results from the Rotterdam Study, both of which showed that CRP is more strongly associated with insulin resistance in women than in men (34). Also, two large North American studies that included only women reported considerably higher relative risks for multivariable-adjusted associations between CRP and type 2 diabetes (3,13) than two other relatively large North American studies that included men and women (6,12). In addition, recently published results from the cross-sectional Health, Aging and Body Composition Study demonstrated a significant sex interaction between levels of CRP and the presence of diabetes, impaired glucose tolerance, or impaired fasting glucose as determined by an oral glucose tolerance test in the age-, race-, and lifestyle-adjusted model and showed that CRP was not associated with diabetes or hyperglycemic status after further adjustment for physiologic risk factors in men but remained highly associated with the outcome in women (35). However, our results are in contrast with observations from the Hoorn Study, where CRP was only associated with diabetes in men but not in women (11) and with results from two Japanese populations, where an equally strong association between CRP and diabetes was seen in men and women (15,16). Furthermore, results of the present study are in contrast with those of the West of Scotland Coronary Prevention Study and the Kuopio Ischemic Heart Disease Risk Factor Study, both of which demonstrated an independent association between CRP and incident type 2 diabetes in men (7,14).

There may be several explanations for these differences. First, chance could have played a role. All four previous studies that separately analyzed men and women (5,11,15,16) were relatively small. The smallest study included only 54 incident cases (25 men and 29 women) (11), and even the largest included only 131 incident cases (67 men and 64 women) (15). Second, different degrees of adjustment might have affected the results. In our study, differences in HRs between men and women became smaller when the number of covariables in the models increased. Third, different ethnic backgrounds could be responsible for some of the diverging results. The two studies that showed significant and similar associations for diabetes in both sexes were conducted in populations of Japanese origin (15,16). However, since the present study, the Hoorn Study (11), the Rotterdam Study (34), and the West of Scotland Coronary Prevention Study (7) were conducted primarily in Caucasians, ethnicity cannot explain all differences. Fourth, different baseline risks of type 2 diabetes could be another factor influencing the results.

Several physiologic conditions might also explain why the association between CRP and incident type 2 diabetes is stronger in women than in men. The main factors are presumably sex differences regarding body composition and systemic sex hormone levels. Markers of inflammation strongly correlate with measures of adiposity, and this association is generally stronger in women than in men, especially for CRP (20). Furthermore, women have a higher percentage of body fat than men (20). Thus, obesity-triggered inflammation could have a greater impact on the development of insulin resistance and type 2 diabetes in women. The second potentially important aspect may involve differences in endogenous sex hormone levels and their sex-specific association with diabetes risk. A recent meta-analysis demonstrated that low testosterone levels in men but high testosterone levels in women are associated with the risk for type 2 diabetes (17). High concentrations of sex hormone–binding globulin (SHBG) are associated with decreased diabetes risk, but SHBG is significantly more protective in women (17). In postmenopausal women, there is evidence that high concentrations of SHBG are associated with lower CRP levels (36,37), whereas one of these studies, which evaluated both CRP and IL-6, did not find a similar association with IL-6 (37). Thus, it seems reasonable to assume that there may be an interaction between systemic concentrations of endogenous sex hormones and proinflammatory mediators regarding diabetes risk, although the present study cannot clarify this issue.

Out of the five studies that have analyzed associations between IL-6 and incident type 2 diabetes (3,9,10,12,13), three included men and women (9,10,12).

Apart from the Gila River Indian Community Study, which was relatively small (10), all studies observed a significant independent association between IL-6 and incident type 2 diabetes. Furthermore, as in the present study, no sex differences were observed in the two studies that found an association and included men and women (9,12).

The strengths of our study include its population-based design, a long follow-up period with a relatively large number of incident cases, inclusion of both sexes, and detailed, standardized assessment of a large number of potential confounding variables. However, several limitations of our study should also be mentioned. First, diabetes diagnoses were initially identified by self-report of the subjects. Although we subsequently validated this information by medical chart review or contacts with the treating physician, we were only able to identify clinically diagnosed cases. Thus, our control group most likely contained some subjects with undiagnosed diabetes. Since this, however, would bias our results toward the null, it cannot explain the positive associations observed in the present study. Since the prevalence of diabetes at baseline was also only determined by self-report, some of the case subjects could have had undiagnosed diabetes at baseline. However, most of these case subjects were probably diagnosed during the first years of follow-up, and exclusion of subjects diagnosed within the first 5 years of follow-up had only a relatively small impact on the observed associations. Second, although we assessed the type of diabetes by contacting the treating physician or by medical chart review and excluded incident cases with a diagnosis of non–type 2 diabetes, some of our cases could, in fact, have had latent autoimmune diabetes in adults instead of type 2 diabetes, since we did not measure GAD antibodies. Third, measures of central obesity such as WHR or waist circumference were only available in a subsample. Therefore, we were not able to adjust for central obesity in the main analyses. However, subgroup analyses demonstrated that the impact of further adjustment for central obesity was relatively small, and, thus, differences in central obesity most likely cannot explain the observed results. Fourth, we did not have any information on menopausal status of women, and, thus, age had to be used as a surrogate for menopausal status. Fifth, selection bias due to missing blood samples or missing follow-up data could, in theory, have influenced the results. However, we have little a priori reason to believe that the association between inflammatory markers and incident type 2 diabetes should be different in subjects without available follow-up data or with missing blood samples. Also, baseline levels of the major cardiovascular risk factors differed only slightly between subjects that had to be excluded and the source population used for the present study. Sixth, markers of inflammation were only measured at a single point in time, at baseline. Thus, we cannot exclude some misclassification of exposure. Finally, samples were stored for a number of years before analysis of inflammatory markers, and it cannot be excluded that this might have led to the degradation of the markers. However, levels of CRP and IL-6 were comparable in all three surveys, although samples had been stored for different periods of time.

In conclusion, we found that elevated levels of CRP and IL-6 are associated with an increased risk of type 2 diabetes in men and women. However, especially for CRP, associations were considerably stronger in women than in men, and, after multivariable adjustment, CRP was only independently associated with risk for type 2 diabetes in women but not in men. Given previously described sex differences in associations between diabetes, obesity, sex hormones, and inflammation, an elucidation of the interaction between sex and inflammation is desirable before CRP and other proinflammatory mediators might be used as prognostic markers.

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