Transforming Growth Factor-β1 and Incident Type 2 Diabetes

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

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OBJECTIVE — Subclinical inflammation leads to insulin resistance and β-cell dysfunction. This study aimed to assess whether levels of circulating transforming growth factor-β1 (TGF-β1)—a central, mainly immunosuppressive, and anti-inflammatory cytokine—were associated with incident type 2 diabetes.

RESEARCH DESIGN AND METHODS — We measured serum levels of TGF-β1 from 460 individuals with and 1,474 individuals without incident type 2 diabetes in a prospective case-cohort study within the population-based MONICA (MONItoring of Trends and Determinants in Cardiovascular Disease)/KORA (Cooperative Health Research in the Region of Augsburg) cohort.

RESULTS — Elevated TGF-β1 concentrations were associated with higher, not lower, risk for type 2 diabetes (age-, sex-, and survey-adjusted hazard ratios [95% CI] for increasing TGF-β1 tertiles: 1.0, 1.08 [0.83–1.42], and 1.41 [1.08–1.83]; \( P \) for trend = 0.012). Adjustment for BMI and metabolic and lifestyle factors had virtually no impact on the effect size.

CONCLUSIONS — Elevated serum concentrations of the cytokine TGF-β1 indicate an increased risk for type 2 diabetes. TGF-β1 may be upregulated to counterbalance metabolic and immunological disturbances preceding type 2 diabetes.

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Subclinical inflammation represents one important mechanism in the development of insulin resistance and β-cell dysfunction, and a systemic proinflammatory state is associated with increased risk for type 2 diabetes (1). However, data on anti-inflammatory immune mediators are scarce. So far, adiponectin remains the only immune mediator for which increased circulating levels indicate a reduced risk for type 2 diabetes and for which a protective effect is biologically plausible (2–4). Transforming growth factor-β1 (TGF-β1) is an interesting candidate in this context because it is a critical regulator of the immune system with mainly immunosuppressive effects (5). In addition to effects on T-cells, TGF-β1 inhibits or reverses the activation of macrophages by interfering with signaling via toll-like receptor–dependent pathways and downregulating central effector mechanisms of the innate immunity such as lipopolysaccharide-induced production of proinflammatory cytokines, reactive oxygen species, and reactive nitrogen species (6,7).

Recent data on interleukin (IL)-1 receptor antagonist (IL-1Ra) from the Whitehall II study led to the hypothesis that the immunological changes before type 2 diabetes do not only include upregulation of proinflammatory mediators but also of anti-inflammatory cytokines—presumably an attempt to counterbalance subclinical inflammation (8). We tested this hypothesis in the large, population-based MONICA (MONItoring of Trends and Determinants in Cardiovascular Disease)/KORA (Cooperative Health Research in the Region of Augsburg) case-cohort study by investigating the association between TGF-β1 serum levels and incident type 2 diabetes.

RESEARCH DESIGN AND METHODS — Data are based on a prospective case-cohort study within the population-based MONICA/KORA cohort (9–11). This study comprises 1,934 participants (255 men/205 women with and 724 men/750 women without incident type 2 diabetes), aged 35–74 years, from a source population of 7,936 study participants (supplementary Fig. A1, available in the online appendix at http://care.diabetesjournals.org/cgi/content/full/dc09-0476/DC1). Mean ± SD follow-up time was 10.9 ± 4.7 years.

The incidence of type 2 diabetes between participants’ study start dates and 31 December 2002 was assessed using a questionnaire sent to all participants of the three baseline surveys (S1: 1984–1985; S2: 1989–1990, and S3: 1994–1995) in 1997–1998 and 2002–2003. Furthermore, all S1 participants were invited to a follow-up examination in 1987–1988. Only subjects for whom the treating physician clearly reported a diagnosis of type 2 diabetes or for whom a diagnosis of type 2 diabetes was mentioned in the medical records or who were taking antidiabetes medication were classified as case subjects.

Further information on study design; collection of demographic, anthropometric, clinical, metabolic, immunological, and lifestyle variables; and the statistical analysis is given in the online appendix.
RESULTS — Baseline characteristics of the study participants have been described for almost identical samples (9–11). Data for the present sample are given in supplementary Table A1. Serum concentrations of TGF-β1 (weighted mean ± SE) were 35.8 ± 0.4 ng/ml in case subjects and 35.2 ± 0.2 ng/ml in noncase subjects (P = 0.16). Given that there was no evidence for interaction between TGF-β1 and sex in the association with type 2 diabetes (P = 0.94 in the age-, sex-, and survey-adjusted model), men and women were analyzed together.

Case subjects were older than noncase subjects, and TGF-β1 levels were negatively correlated with age (supplementary Table A2). Adjustment for age only resulted in hazard ratios (HRs) (95% CI) for increasing TGF-β1 tertiles of 1.0, 1.08 (0.82–1.40), and 1.33 (1.03–1.73) (P_{trend} = 0.024). This association was slightly stronger when we also adjusted for sex and survey (model 1, Table 1) and remained almost unchanged when we additionally adjusted for BMI and lifestyle factors (smoking status, alcohol consumption, and physical activity) in model 2 and for systolic blood pressure, total/HDL cholesterol, and parental history of diabetes (negative, positive, and unknown); model 4 adjusted for factors in model 3 plus C-reactive protein, MIF, IL-8, soluble E-selectin, and RANTES (all included in the models stratified in sex-specific tertiles; sample size after exclusion of subjects with missing values for additional biomarkers: n = 1,947).

CONCLUSIONS — This study found that elevated serum concentrations of TGF-β1 are associated with incident type 2 diabetes. The associations remained stable in multivariate analyses taking into account demographic, anthropometric, metabolic, and lifestyle factors. Our data substantiate and extend our recent report on IL-1Ra and incident type 2 diabetes in the Whitehall II study (8). Although IL-1Ra is an anti-inflammatory cytokine that improves metabolic control in patients with type 2 diabetes (12), we found that elevated, not decreased, levels of IL-1Ra preceded the development of type 2 diabetes (8). This new finding underlines that the immune activation during the development of type 2 diabetes is complex and includes both pro- and anti-inflammatory mediators. One explanation could be that elevated concentrations of TGF-β1 and IL-1Ra represent a counter-regulation of the proinflammatory state that increases the risk for type 2 diabetes.

As an alternative explanation, TGF-β1 could also play a proinflammatory role in the development of type 2 diabetes. The action of TGF-β1 depends on the microenvironment (different leukocyte subsets and cytokines), and TGF-β1 can also positively regulate immune responses (13). In the presence of IL-6, TGF-β1 supports the differentiation of T-helper 17 (Th17) cells that are activated in many proinflammatory conditions. Interestingly, IL-6 levels were also increased before the onset of type 2 diabetes in our study (14). Activated Th17 cells release proinflammatory cytokines such as IL-17 that has been reported to stimulate nitric oxide–mediated β-cell toxicity in a mouse model (15).

The large sample size, the population-based study design, and the long follow-up period are strengths of our study. Regarding the limitations, we measured total TGF-β1 after acidification of sera and release of latent TGF-β1 instead of biologically active TGF-β1. However, active TGF-β1 has a half-life of 2 min so that its quantification was not feasible.

In conclusion, elevated serum concentrations of the mainly immunosuppressive cytokine TGF-β1 precede the manifestation of type 2 diabetes. Further studies are needed to elucidate whether upregulation of TGF-β1 represents an attempt of a protective response against yet unrecognized metabolic and immunological disturbances during the development of type 2 diabetes or whether elevated TGF-β1 levels in the circulation may directly contribute to mechanisms that favor insulin resistance, β-cell dysfunction, and type 2 diabetes.

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No potential conflicts of interest relevant to this article were reported.

Parts of this study will be presented in abstract form at the 45th annual meeting of the European Association for the Study of Diabetes, Vienna, Austria, September 29–October 2009.

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