Inflammatory Markers and Diabetic Retinopathy in Type 1 Diabetes

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Retinopathy affects ~86% of people in the U.S. with type 1 diabetes (1). This percentage is higher than would be expected as a result of poor glycemic control, blood pressure elevations, and smoking. Another possible risk factor for microvascular disease in type 1 diabetes might be inflammation. Inflammation, as measured by C-reactive protein (CRP), has been shown to be increased in people with type 1 and type 2 diabetes who have macrovascular complications (2-5). A recent study in laboratory animals has suggested that inflammation may be important in the etiology of diabetic retinopathy (6). The purpose of this study was to determine if there was an association between inflammatory markers and diabetic retinopathy.

RESEARCH DESIGN AND METHODS — This was a cross-sectional prospective study comprised of subjects with type 1 diabetes (n = 154) matched by age and sex with nondiabetic control subjects in a 3:1 ratio. Eligible subjects (control subjects and type 1 diabetic patients) were between the ages of 14 and 42 years. Test subjects had type 1 diabetes for >5 years. Subjects were excluded if they had a blood pressure >140/90 mmHg, were tobacco users, or were on medications known to affect CRP (ACE inhibitors, hydroxymethylglutaryl CoA reductase inhibitors, nonsteroidal anti-inflammatory agents, and aspirin), had other known concomitant illnesses, or were pregnant. Written informed consent was obtained from willing participants using institutional review board–approved consent forms and Health Insurance Portability and Accountability Act–compliant authorization forms.

Eye examinations included direct ophthalmoscopy (with pupils dilated) as well as digital retinal photographs that were obtained with a Canon CR6-45NM D60 camera. Three fields were photographed for each eye: optic nerve and macula, nasal retina, and temporal retina. A single ophthalmologist (W.E.J.) did the grading for the eye exams using the modified Airlie House classification (7).

All samples were spun in a refrigerated centrifuge at 2,000 ×g for 10 min at 4°C. Plasma and serum were extracted and stored at −70°C until assayed. The plasma samples for prostaglandin E1 (PGE1) and prostaglandin E2 (PGE2) were stored in special tubes containing 100 µg indomethacin. The HbA1C was measured with the DCA-2000 Meter (Bayer Laboratories, Elkhart, IN). IL-6, PGE1, and PGE2 were assayed using colorimetric sandwich enzyme-linked immunosorbent assay kits from R&D Systems (Minneapolis, MN).

RESULTS — There were a total of 115 type 1 diabetic subjects (51 males and 64 females) with a mean ± SD age and duration of diabetes of 26.7 ± 7.2 and 16.0 ± 7.9 years, respectively. Thirty-nine control subjects (18 males and 21 females) with a mean ± SD age of 26.6 ± 6.7 years were enrolled. Age (P = 0.001), BMI (P = 0.02), and duration of diabetes (P = 0.001) were significantly higher in the 39 diabetic subjects with grades 4–6 retinopathy compared with subjects with grades 2/3 retinopathy (42 subjects) or no retinopathy (37 subjects). There were no significant differences in HbA1C levels (7.8–8.0%) among the three groups (P = 0.79).

PGE1 levels were significantly higher in the diabetic population than in the control group (P < 0.001) (Table 1). This was true for all levels of retinopathy. This relationship remained significant after controlling for the effects of BMI. All other markers of inflammation were similar between the two groups.

A significant relationship was found (P = 0.01) between grades of retinopathy and CRP. However, after controlling for age, duration of diabetes, sex, and BMI, the significance of the relationship was lost (P = 0.42). None of the other markers of inflammation were significantly associated with retinopathy.

For the diabetic population, BMI was
positively correlated with CRP and IL-6 before adjustment for age, sex, and duration of diabetes (P < 0.001 for both) and after adjustment (P = 0.04 and 0.02, respectively). Age (P = 0.002), BMI (P = 0.001), and IL-6 (P = 0.001) were the only significant predictors of CRP. The relative increase in CRP for each of these variables was adjusted for the other variables in the model. IL-6 and CRP were also positively correlated with each other.

CONCLUSIONS — The purpose of this study was to determine the contribution of inflammation to the pathogenesis of retinopathy in subjects with type 1 diabetes. Although CRP was significantly associated with the eye grades, the significance was lost after controlling for the effect of other covariables (age, duration of diabetes, and BMI). Other markers of inflammation (IL-6, PgE1, and PgE2) did not vary significantly among groups of type 1 diabetic subjects classified according to the severity of retinopathy. Kilpatrick et al. (9) used multivariate analyses to show age, BMI, and HbA1c to be associated with higher CRP concentrations in adults with type 1 diabetes. Although CRP was significantly associated with albumin excretion using univariate analysis, the association was lost using multiple regression.

As described previously (10), PgE2 values for the type 1 diabetic subjects were consistently higher than for the control subjects. The PgE2 levels, however, did not correlate with retinal grade. PgE2 is believed to be involved in inflammation, apoptosis, angiogenesis, and increasing vascular permeability (11). The consistent elevation of PgE2 in the diabetic population found in this study raises the possibility that this may be a factor in the etiology of microvascular complications.

In summary, this cross-sectional study of inflammatory markers and the retinal complications of type 1 diabetes showed that PgE2 was the only marker significantly elevated in diabetic subjects compared with control subjects. However, PgE2 was not significantly related to varying degrees of retinopathy among the diabetic subjects.

Acknowledgements — These findings were presented in part at the American Diabetes Association 64th annual meeting in Orlando, Florida, 4–8 June 2004.

References

Table 1—Levels of inflammatory markers in diabetic and control subjects

<table>
<thead>
<tr>
<th></th>
<th>log CRP</th>
<th>log IL-6</th>
<th>log PgE1</th>
<th>log PgE2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diabetic subjects (n = 154)</td>
<td>0.10 ± 0.55</td>
<td>0.05 ± 0.22</td>
<td>2.60 ± 0.29</td>
<td>2.41 ± 0.28</td>
</tr>
<tr>
<td>Control subjects (n = 39)</td>
<td>−0.05 ± 0.51</td>
<td>−0.01 ± 0.29</td>
<td>2.55 ± 0.20</td>
<td>2.03 ± 0.37</td>
</tr>
<tr>
<td>P</td>
<td>0.14</td>
<td>0.21</td>
<td>0.26</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Data are means ± 1 SD of the logs of the assay values.