Serum vascular endothelial growth factor in late rheumatoid arthritis

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ABSTRACT

Objectives
To investigate the serum levels of VEGF in patients with rheumatoid arthritis of long duration.

Methods
Serum VEGF levels were measured in 118 patients with long-standing rheumatoid arthritis according to the ACR criteria (mean duration 12 years). The disease activity score was evaluated by the method of van der Heijde et al.

Results
Serum levels of VEGF in patients with RA were significantly higher than in healthy controls. VEGF levels showed no correlation with CRP, SAA amyloid protein, or the disease activity score.

Conclusions
Our findings suggest that, contrary to the results reported in patients with early onset RA, where VEGF appears to play an active part in joint inflammation, in long-standing RA elevated VEGF serum levels may be an independent marker although its significance remain to be established.

Introduction
Rheumatoid arthritis (RA) is a chronic debilitating disease characterized by distinct autoimmune, inflammatory and fibrovascular components which lead to synovial proliferation and joint destruction. Neovascularization of the inflamed synovium is a hallmark of RA. Angiogenesis appears to contribute to synovial growth, thus potentiating disease progression (1, 2).

Neovascularization is a complex process involving endothelial cell division, selective degradation of vascular basement membrane and endothelial cell migration (3). A number of angiogenic factors may be important in the neovascularization found in the RA joint. One of them is vascular endothelial growth factor (VEGF), endothelial cell-specific mitogen in vitro and an angiogenic growth factor in vivo. VEGF have been reported to play an important role in human RA and experimental models (4, 5).

Serum levels of VEGF have been reported to be elevated in patients with RA and to correlate with disease activity and inflammatory markers. In neither experimental models nor in humans has the time course of serum VEGF levels and its corresponding overexpression been fully explored and the suggestion was recently made that these correlations are only seen in RA of early to median disease duration, where the inflammatory events are more pronounced (6-8).

In the present study we examined serum VEGF levels in patients with long-standing RA and correlated the findings with inflammatory markers and disease activity indexes.

Material and methods

Patients
118 patients with long-standing rheumatoid arthritis according to the ACR criteria attending the outpatient clinic of the University Hospital from the State University of Rio de Janeiro were studied: 106 women and 12 men with a mean age of 51.6 years and a mean duration of 12 years (Table I). The disease activity score (DAS) was evaluated by the method of van der Heijde D and co-workers (9). Only 10 patients had non-erosive disease. According to the Eular criteria none of our patients were in remission or and burned out disease (DAS <1). The drugs employed in the treatment of these patients are shown on Table II.

Analysis
The laboratory inflammatory markers used were serum levels of C reactive protein (CRP) and serum amyloid (SAA). Both assays were performed as previously described (10). Vascular endothelial growth factor (VEGF) serum levels were quantified using a sensitive sandwich Elisa (RD Systems, Minnesota, USA).

Table I. Characteristics of the patients and controls.

<table>
<thead>
<tr>
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<th>Patients</th>
<th>Controls</th>
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<tbody>
<tr>
<td>Number</td>
<td>118</td>
<td>50</td>
</tr>
<tr>
<td>Disease duration (years)</td>
<td>12.0</td>
<td>-</td>
</tr>
<tr>
<td>Mean age (years)</td>
<td>51.6</td>
<td>42.0</td>
</tr>
<tr>
<td>Disease score (range)</td>
<td>2.5 - 7.0</td>
<td>-</td>
</tr>
<tr>
<td>CRP (mg/dL) (range)</td>
<td>3 - 15</td>
<td>0.2 - 0.8</td>
</tr>
<tr>
<td>SAA (mg/ml) (range)</td>
<td>3 - 800</td>
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Statistical analysis
Spearman’s rank test and a linear multiple regression analysis were employed in our study.

Results
Patients with RA had significantly higher levels of VEGF when compared to normal individuals, 335 ± 86 versus 188 ± 133 pMol/L (Table I) (p = 0.01). The correlation coefficient of VEGF with CRP and SAA were below 0.2 (Figs. 1 and 2). When we employed the Eular disease activity score (DAS) criteria and tested for correlations with VEGF they were also low (Fig. 3). We also tested for correlations between DAS and CRP and SAA and found low to moderate values 0.53 and 0.49 respectively, both significant (pp = 0.01). Correlations between CRP and SAA showed the best results r of 0.7 (pp = 0.001) (data not shown). No significant differences were observed between the presence of erosions and VEGF levels or between VEGF X DAS in the presence or absence of elevated CRP and SAA.

Discussion
As mentioned in the Results section and in previously published papers, inflammatory markers do show some correlation with disease activity in patients with rheumatoid arthritis of long duration (11). In the current study our results show that VEGF serum levels are elevated in patients with late RA similar to the results obtained in early onset RA and JRA (8,12). The fact that we could not find correlations with inflammatory markers is somewhat surprising and points to mechanisms different from those suggested in early RA for the sustained VEGF elevation. It appears that the elevations reported in juvenile rheumatoid arthritis and in experimental models are related to cytokine-mediated pathways associated with angiogenesis as the main
mechanism of elevated VEGF levels. In man, more advanced disease is usually associated with symptoms derived from chronic debilitating joint destruction where pain comes more from mechanical distress than the presence of active inflammatory disease. Therefore, it is not surprising that in this patient population we could not find a good correlation between VEGF levels, DAS and acute phase reactants. In fact in a recently published study, the administration of anti-serum against VEGF blocked induced neovascularization - a critical step in the early development of arthritis, but severity was unaffected when the antiserum was used after the onset of the disease (late stages) (5).

On the other hand, the reasons for sustained elevations of VEGF not associated with inflammation are not completely understood at this time. Our data shows that increased serum VEGF concentrations in patients with late RA may be an independent marker, somewhat similar to what is seen in other inflammatory and neoplastic diseases (13, 14).

Further studies are needed to contribute to our understanding of the role of VEGF in late stages of RA. The concomitant measurement of other angiogenic markers may shed light on this matter and are being planned by our laboratory.

References