Examination of HLA-DR4 as a severity marker for rheumatoid arthritis in Greek patients

Kyriaki A Boki, Alexandros A Drosos, Athanasios G Tzioufas, Jerry S Lanchbury, Gabriel S Panayi, Haralampos M Moutsopoulos

Abstract

Objectives—Previous reports have shown that HLA-DR4 may be a severity marker for rheumatoid arthritis (RA) in patients of northern European origin. The aim of the present study was to investigate this relation in Greek patients with RA, as RA in Greece differs from the RA described previously on clinical, serological, and immunological grounds.

Methods—Eighty four patients were studied in whom HLA-DR typing was performed by restriction fragment length polymorphism and the subtypes of HLA-DR4 were determined by the polymerase chain reaction. The absence or presence of HLA-DR4 and its subtypes was correlated with the clinical and serological characteristics of the patients and with the side effects due to disease modifying drugs.

Results—Twenty one of the 84 (25%) patients with RA were DR4+. There was no difference between the DR4+ and DR4− patients with respect to duration of disease, severity of arthritis, functional grade, and joint erosion score. The DR4+ group were more likely to have side effects due to disease modifying drugs (43%) than DR4− patients (36%), but this difference was not statistically significant. DR4− patients had more extra-articular manifestations, including Sjögren’s syndrome (47 v 19%). Analysis of the DR4 subtypes showed that Dw15 was the most common variant (9/21 patients; 43%). There was no statistical difference in the clinical manifestations among patients with different DR4 subtypes. The same was also true when the clinical picture was correlated with the ‘shared RA epitope’ (QKRAA/QRRRA/RRRAA), which is common to all HLA-DRB1 alleles positively associated with RA.

Conclusions—These results suggest that HLA-DR4 is not a severity marker in Greek patients with RA and further indicate differences in the clinical expression of RA in Greece.
had more side effects due to disease modifying drugs (43%) than the DR4 negative patients (36%), but this difference was not statistically significant.

Dw15 was the most common variant and was observed in nine patients (43% of DR4 positive patients). The other DR4 subtypes (Dw4, Dw14, Dw13) were also detected but the numbers were too small for statistical analysis. An interesting observation was that all the Dw4, Dw14, and Dw13 patients were seropositive for rheumatoid factor. There was no difference in clinical manifestations among patients with different DR4 subtypes. The Dw10 variant was not detected in our patients with RA. The HLA-DRβ1 chains of Dw4, Dw14, and Dw15 variants of DR4, DR1, and Dw10 share a similar third hypervariable region amino acid sequence (QKRAA/QRRAA/RRRAA on the single letter amino acid code), which is called 'the shared epitope'.

When the clinical data for the patients with the shared RA epitope was compared with the rest of the patients with RA, no statistical difference was observed. The Dw16 variant of Dw6 has a similar sequence (QRAA) but it was not detected in our patients.

Table 1: Rheumatoid arthritis in HLA-DR4 positive and HLA-DR4 negative Greek patients

<table>
<thead>
<tr>
<th>Clinical findings</th>
<th>HLA-DR4+ (n=21)</th>
<th>HLA-DR4- (n=63)</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Female/male</td>
<td>17/4</td>
<td>54/9</td>
<td></td>
</tr>
<tr>
<td>Mean (SD) age (years)</td>
<td>58.2 (12.5)</td>
<td>54.1 (11.1)</td>
<td>NS</td>
</tr>
<tr>
<td>Mean (SD) disease duration (years)</td>
<td>11.0 (8.9)</td>
<td>9.6 (6.3)</td>
<td>NS</td>
</tr>
<tr>
<td>Seropositivity (%)</td>
<td>85</td>
<td>73</td>
<td></td>
</tr>
<tr>
<td>No with antibodies to Ro (SSA)</td>
<td>4</td>
<td>7</td>
<td>NS</td>
</tr>
<tr>
<td>Mean (SD) No of active joints</td>
<td>5.7 (4.2)</td>
<td>6.2 (5.6)</td>
<td>NS</td>
</tr>
<tr>
<td>No with extra-articular features*</td>
<td>1</td>
<td>12</td>
<td>NS</td>
</tr>
<tr>
<td>No with Sjögren’s syndrome</td>
<td>3</td>
<td>18</td>
<td>NS</td>
</tr>
<tr>
<td>No with EMSS†</td>
<td>4</td>
<td>30</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>No with side effects‡</td>
<td>9</td>
<td>23</td>
<td>NS</td>
</tr>
</tbody>
</table>

NS=not significant.
*Rheumatoid nodules, Raynaud’s phenomenon, serositis, skin vasculitis, livedo reticularis, and lymphadenopathy.
†Extra-articular features and Sjögren’s syndrome.
‡Side effects due to sodium aurothiomalate or d-penicillamine (rash, leucopenia, thrombocytopenia, proteinuria).

Table 2: Hand radiographs scored according to the Steinerheuer classification in Greek patients with RA who were HLA-DR4 positive and HLA-DR4 negative

<table>
<thead>
<tr>
<th>Grade</th>
<th>Percentage of radiographs positive</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>HLA-DR4+</td>
</tr>
<tr>
<td>I</td>
<td>18</td>
</tr>
<tr>
<td>II</td>
<td>38</td>
</tr>
<tr>
<td>III</td>
<td>19</td>
</tr>
<tr>
<td>IV</td>
<td>5</td>
</tr>
</tbody>
</table>

Discussion
Rheumatoid arthritis can present with a wide clinical spectrum of expression from mild to severe articular disease with or without extra-articular manifestations. It is associated with HLA-DR4 or DR1 and molecular analysis of the third hypervariable region of these molecules has revealed a similar molecular epitope which has substantiated the ‘shared epitope’ hypothesis first proposed by Gregersen et al. The implications for the presentation of unknown rheumatoid antigens to T cells is obvious. Since the discovery of the association of RA with HLA-DR4, various workers have attempted to define prognostic markers based on the presence or absence of HLA-DR4; some workers have suggested that patients with RA who are HLA-DR4 positive have more severe articular disease associated with extra-articular manifestations, so that, for example, 96–100% of patients with RA and Felt’s syndrome are DR4 positive, whereas other workers have not found these associations.

Rheumatoid arthritis in Greece is a milder joint disorder than RA in England, with significantly fewer extra-articular manifestations and an almost total absence of Felt’s syndrome. These clinical differences may be partly based on immunogenetic differences as previously described by serological typing and extended by the present molecular analysis in which 43% of Greek patients with RA were shown to possess the disease susceptibility hypervariable region compared with British patients in whom it was present in over 80%. In the present study, evaluation of the articular and extra-articular manifestations of Greek patients with RA with or without HLA-DR4 or its subtypes did not reveal any
significant differences. The observation that Greek patients with RA with extra-articular manifestations belong to the DR4 negative group is due mainly to Sjögren’s syndrome, as analysis of the extra-articular features of the patients excluding Sjögren’s syndrome yielded no statistical differences between patients with RA with and without HLA-DR4 or its subtypes. Also of interest is the observation that all Greek patients with RA with the HLA-DR4 subtypes Dw4, Dw14, or Dw13 were seropositive for rheumatoid factor. Dw4 and Dw14 variants have previously been associated with seropositive RA. The mechanisms by which these DR4 Dw subtypes influence rheumatoid factor production is not known. Rheumatoid factor seropositivity in most studies, however, has been associated with severe erosive disease and the presence of extra-articular features, so the primary association is unclear.

One way in which Greek patients with RA differ significantly from those of northern Europe is in the distribution of DR4 subtypes among DR4 positive patients. Rheumatoid arthritis and, particularly, Felty’s syndrome, are most strongly associated with the HLA-Dw4 subtype, which is much less common among DR4 positive Greek patients. A study of southern Chinese patients with RA has reported that although Dw14 and Dw15 were the most common subtypes among DR4 positive subjects, Dw4 was lacking and these patients showed relatively milder disease than that commonly encountered in northern Europe. This situation is similar to that in our Greek patients and may reflect the fact that the Dw4 subtype is the main determinant of disease severity in a population. Given its rarity in Greek patients, further large scale studies will be necessary to evaluate this hypothesis.

To explain the genetic heterogeneity among Greek patients with RA the ‘shared RA epitope’ was also investigated. It was found that only 43% of the patients had this epitope. This finding is in contrast with previous studies, where each of the major susceptibility genes associated with RA in DR4 positive and DR4 negative subjects share related amino acid sequences. Hence, this finding implies that the ‘shared epitope’ does not seem to play as crucial a part in the aetiopathogenesis of Greek patients with RA.

These observations further support the proposal that RA in Greek patients differs from RA reported previously mainly in patients of northern European origin. The clinical similarities of the Greek patients with RA with or without DR4 or its subtypes raise important questions about the role of these molecules in the initiation and perpetuation of autoimmune lesions of RA in Greece. One conclusion may be that DR4, DR1, and DR10 positivity, in this group, is a susceptibility rather than a severity marker. Furthermore, the immunogenetic and clinical features observed in Greek patients with RA suggest that other gene(s) in or outside the HLA region or environmental factors, or both, may be influencing or modifying disease expression.

We thank the Welcome Trust for an inter-laboratory collaborative grant to JSL, GSP, and HMA and the Arthritis and Rheumatism Council (grants U9 and P75, and L45).

Examination of HLA-DR4 as a severity marker for rheumatoid arthritis in Greek patients.
K A Boki, A A Drosos, A G Tzioufas, J S Lanchbury, G S Panayi and H M Moutsopoulos

*Ann Rheum Dis* 1993 52: 517-519
doi: 10.1136/ard.52.7.517

Updated information and services can be found at:
http://ard.bmj.com/content/52/7/517

**Email alerting service**

*These include:*

Receive free email alerts when new articles cite this article. Sign up in the box at the top right corner of the online article.

**Notes**

To request permissions go to:
http://group.bmj.com/group/rights-licensing/permissions

To order reprints go to:
http://journals.bmj.com/cgi/reprintform

To subscribe to BMJ go to:
http://group.bmj.com/subscribe/