HLA and rheumatoid arthritis: a combined analysis of 440 British patients


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Summary

Four hundred and forty unrelated British Caucasoid patients with rheumatoid arthritis (RA) have been HLA typed for class I and class II antigens. Analyses of HLA antigen associations were performed on the overall group and in patient subsets selected according to particular disease parameters or sex, or both. The results confirm previously reported positive associations of HLA-DR4, Dw4, and DRw53 and negative associations of HLA-DR2 and DR7 with RA. Patients subsets with severe erosions, seropositivity, and features of extra-articular disease showed a stronger association, also confirming earlier reports. The link between HLA and disease severity was emphasised by a significant trend of increased Dw4 frequency with increasing severity of radiological erosions. In addition, a positive association of RA with HLA-A2 was observed and a strong negative association with DR3. The frequency of HLA-B27 was significantly increased in patients with subluxation of the spine. Differences were observed between male and female patients in relation to the HLA association. In men an increase in the frequency of the haplotype HLA/Dw4/DR4/Bw62/Cw3/A2 was observed. This showed no relationship with parameters of disease severity other than extra-articular disease. In women only class II antigens (DRw53/Dw4/DR4) showed an increased frequency. This increase was strongly associated with disease severity. A significant decrease of this class II association was observed with increasing age of disease onset; this was not seen in men.

Key words: HLA antigen associations, disease severity, seropositivity, erosions, subluxation of the spine, extra-articular disease, age of onset.

Rheumatoid arthritis (RA) is a relatively common condition, of which the aetiology remains unknown and the basis for susceptibility in individuals is still unclear. A major advance in our understanding of RA development came with the observation that certain HLA antigens were strongly associated with this disease. These antigens include HLA-Dw4 and DR4, and DRw53 (MT3). No association with HLA-A or B locus antigens was found in the early studies, though several later reports suggested other HLA antigen associations.

In most of the reported studies the frequency of DR4 in RA patients is within the range 50–75%, whereas the frequency of DR4 in controls ranges between 25% and 35%. Such association argues for the existence of an RA predisposing factor which is cotransmitted (linked) or associated with HLA. This factor could be DR4 itself or a component of susceptibility in linkage disequilibrium with HLA-DR4. These findings must be interpreted in the context of the known familial tendency to develop RA. In the number of small family studies which have been performed, however, the evidence for genetic linkage between HLA and RA remains weak. It is likely that the susceptibility to and development of RA are not solely due to one predisposing factor but, rather, are polygenic and may include environmental factors. On the other hand, the apparent heterogeneity of the disease suggests that what is presently designated as rheumatoid arthritis may represent more than one

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nosological entity. The clarification of the relation between all components involved in the disease, including HLA, could help to resolve this question.

In the studies of HLA and disease three observations must be made in relation to the interpretation of any results. The first is that the patient sample size can limit the confidence placed on any observation. Secondly, most reports of adequate sample size have usually been from international collaborative studies involving heterogeneous HLA results from different laboratories. The third is that any comparisons require homogeneity of ethnic origin between both patient and control panels. This is important because of the varying distribution of HLA polymorphisms in different populations. This lack of homogeneity may be a valid criticism of a number of international collaborative studies. In this report attempts to overcome these three problems have been made.

In the past six years a total of 440 Caucasoid RA patients of British ancestry (British Caucasoids) have been HLA typed in the Department of Immunology of The London Hospital Medical College. The patients were attending different centres but were all part of various collaborative studies of HLA and RA. This large sample of RA patients is homogeneous with respect to both ethnic origin of the population studied and the quality control of all the HLA typing. We have now pooled all these data in order to re-examine several findings reported before by ourselves and other groups of investigators.

Patients and methods

Patients

Four hundred and forty unrelated British Caucasoid RA patients from several groups have been studied. The patient selection criteria and the distribution of these patients in the different study groups are shown in Table 1. All patients were classified according to the criteria of the American Rheumatism Association (ARA). Of the patients studied, 153 fulfilled the ARA criteria for classical RA, 233 had definite RA, and 50 probable or possible RA. Confirmed classifications were not available for four patients.

Controls

The control panel consisted of 108 British Caucasoid

Table 1 Description of patients in the different groups studied

<table>
<thead>
<tr>
<th>Group</th>
<th>n</th>
<th>M</th>
<th>F</th>
<th>Centre</th>
<th>Criteria of selection</th>
<th>Duration (years)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bath</td>
<td>52</td>
<td>16</td>
<td>36</td>
<td>Royal National Hospital for Rheumatic Disease, Bath</td>
<td>Prospective study of classical or definite RA patients within one year of onset</td>
<td>&gt; 25</td>
<td>34, 43, 44</td>
</tr>
<tr>
<td>BJRU</td>
<td>63</td>
<td>21</td>
<td>42</td>
<td>Bone and Joint Research Unit, The London Hospital</td>
<td>Prospective study of early diagnosed RA patients of any ARA* category included</td>
<td>1–3</td>
<td>45</td>
</tr>
<tr>
<td>Kennedy</td>
<td>78</td>
<td>15</td>
<td>63</td>
<td>Kennedy Institute for Rheumatology</td>
<td>Retrospective study of classical (71) or definite (seven) RA patients, of which 27 had extra-articular disease</td>
<td>1–18</td>
<td>28</td>
</tr>
<tr>
<td>London</td>
<td>50</td>
<td>12</td>
<td>38</td>
<td>Department of Rheumatology, The London Hospital</td>
<td>Retrospective study of probands with classical or definite RA, part of family study</td>
<td>3–15</td>
<td>37</td>
</tr>
<tr>
<td>MALES</td>
<td>15</td>
<td>15</td>
<td>0</td>
<td>Department of Rheumatology The London Hospital</td>
<td>Retrospective study of male patients with classical RA</td>
<td>3–30</td>
<td>—</td>
</tr>
<tr>
<td>RAPS</td>
<td>129</td>
<td>45</td>
<td>84</td>
<td>Depts of Immunology and Rheumatology, Middlesex Hospital</td>
<td>Prospective study of early diagnosed RA patients of all ARA categories</td>
<td>1–15</td>
<td>21, 24, 25</td>
</tr>
<tr>
<td>Taplow</td>
<td>31</td>
<td>9</td>
<td>22</td>
<td>Canadian Red Cross Clinic, Taplow, Berks</td>
<td>Retrospective study of all classical or definite erosive seropositive RA patients</td>
<td>1–28</td>
<td>24</td>
</tr>
<tr>
<td>8th IHW*</td>
<td>22</td>
<td>4</td>
<td>18</td>
<td>Department of Rheumatology, Middlesex Hospital</td>
<td>Retrospective study of classical or definite RA patients, part of the 8th International Workshop</td>
<td>8–15</td>
<td>30</td>
</tr>
</tbody>
</table>

Total 440 137 303

*IHW=International Histocompatibility Workshop; ARA=American Rheumatism Association.
voluntary blood donors who had been HLA-A, B, C, DR, and D typed at the Department of Immunology, The London Hospital Medical College.

**HLA typing**

HLA-A, B, and C typing was performed by a modification of the NIH technique. HLA-DR typing was done by a double fluorescence technique based on that reported by van Rood et al. Local and International Workshop sera were used to detect all defined HLA-A, B, C, and DR specificities.

HLA-D typing was performed as previously described. Double normalised values were calculated by the method of Mendel et al. Antigen assignments for Dw1 to 10 (and Dw13 and 14 in some patients) were based on criteria described elsewhere.

**Clinical and laboratory data**

**Rheumatoid factors**

The results of the conventional latex and RAHA tests were used. Patients were considered seropositive if their latex titre was equal to or more than 1/80 or their RAHA titre was equal or more than 1/40, or both. These tests were performed in the routine laboratories of the different centres involved (see Table 1) and were not standardised.

**Radiological changes**

Sequential radiographs of the hands and feet and of the cervical spine were assessed for the presence and severity of erosions of the hands and feet as described by Lawrence and of atlantoaxial and subaxial subluxations of the cervical spine as described by Bland. The films from the patients assessed for radiological change were read by the same clinician and confirmed by one other clinician as described in a previous report.

**Extra-articular manifestations**

The patients were examined in the different centres for the presence of extra-articular disease, digital or cutaneous vasculitis, fibrosing alveolitis, and neuropathy. The presence of subcutaneous nodules was examined separately.

**Statistical analysis**

HLA antigen frequencies (AF) in the RA panel were assessed for significant deviations from control frequencies by means of a $\chi^2$ analysis. The strength of association was estimated by calculating relative risks (RR) using the formula of Woolf as modified by Haldane. The aetiological (EF) and preventive (PF) fractions were calculated from the RR as
described by Green.\(^1\) When RR > 1 the aetiological fraction estimates which portion of the disease cases is attributable to the antigen out of the total number of cases with the antigen in the total disease population. When RR < 1 the preventive fraction is calculated as a measure of the proportion of cases prevented from developing the disease by the absence (or lower frequency) of the antigen, out of the hypothetical number of individuals who would have developed the disease had the antigen been present at the control frequency. In this report, unless stated otherwise, only previously described associations were tested for statistical significance and thus the p values were not adjusted for the effect of multiple testings. p Values are abbreviated as given in Table 2.

The relation between the frequency of specific antigens and indices of disease severity which could be expressed on an ordinal scale were analysed by Kendall’s tau (\(\tau\)), which gives an indication of linear trend between two variables.\(^2\)

The effect of age at onset was described by examination of 10 year moving averages; the raw data (of presence or absence of specific antigen) were examined for linearity by regression techniques. The parameter b (linear regression coefficient) was calculated.

**Results**

**HLA antigens in each patient group and in the overall RA panel.**

Table 2 shows the HLA antigens with significant positive or negative associations with RA in the total panel of 440 patients and in each of the single groups of patients studied. The frequencies of HLA-DR4, Dw4, and DRw53 were significantly increased. The relative risk (RR) and the aetiological fraction (EF) were greater for DR4 (RR=4.09, EF=0.49) than for Dw4 (RR=2.8, EF=0.31) and DRw53 (RR=2.0, EF=0.35).

Other HLA-DR antigens showed significantly lower frequencies in the RA patients compared with controls: DR2 (RR=0.54\(^*\), EF=0.12), DR3 (RR=0.38\(^***\), EF=0.21), and DR7 (RR=0.43\(^**\), EF=0.16).

The class I antigens with significantly different frequencies in the RA patients compared with the control panel were A2, A1, and B18. HLA-A2 was significantly increased (RR=1.64\(^*\), EF=0.23), and A1 and B18 were found with lower frequencies (RR=0.60\(^*\), EF=0.14 and RR=0.31\(^**\), EF=0.08 respectively).

**HLA association with RA in male and female patients.**

The overall frequency of DR4 in male and female patients was very similar (64.8% in men and 65.6% in women) (Table 3). There were some differences for other antigens, however: the frequencies of HLA-A2, Cw3, and Bw62 were significantly increased in male patients but not in the female RA patients compared with controls. No significant difference in HLA antigen frequency was seen between male and female controls.

**HLA and age of onset of RA**

The proportion of patients with DR4 was analysed by age at disease onset for male and female patients. Results are presented in Fig. 1 as 10 year moving averages to allow for the small numbers at each individual age; i.e., the age of onset is represented in the horizontal axis as a grouped continuous variable, each point representing a 10 year interval. The vertical axis represents the antigen frequency of DR4 for each group. The data show that in female RA patients there is a trend of decline in the frequency of DR4 with advancing age of disease onset. This trend is not apparent in men. The raw data were tested for linearity by linear regression analysis. This confirmed no relation in men and a significant trend in women (b=0.35\%, p<0.01).

**HLA and seropositivity**

Data on seropositivity were available for 409 patients, of which 326 (79.7\%) were seropositive.

| Table 3 HLA antigens in male and female RA patients |
|-----------------------------------------------|-----------------------------------------------|
| **HLA antigen** | **Non-RA controls** | **RA men (n=137)** | **RA women (n=303)** |
| A2 | 47.2 | 71.6 | 2.79 | 0.45 | 54.4 | — | — | NS |
| Cw3 | 26.0 | 39.1 | 1.81 | 0.18 | 32.9 | — | — | NS |
| Bw62 | 8.3 | 27.5 | 2.14 | 0.15 | 16.8 | — | — | NS |
| Dw4 | 25.2 | 44.8 | 2.38 | 0.25 | 50.4 | 2.98 | 0.33 | NS |
| DR4 | 31.3 | 64.8 | 3.97 | 0.48 | 65.6 | 4.14 | 0.50 | *** |
| DRw53 | 54.9 | 64.8 | — | — | 73.8 | 2.30 | 0.42 | *** |

\(\text{AF}=\text{antigen frequency}; \text{RR}=\text{relative risk}; \text{EF/PF}=\text{aetiological/preventive fraction (as in Table 2)}; \text{p}=\text{probability (as in Table 2)}.\)
Fig. 1 The age of disease onset is represented in the horizontal axis as a grouped continuous variable, each point representing a 10 year interval, as follows: 16–25, 17–26, 18–27, etc. In the female patients there is a trend of decline in the frequency of DR4 with advancing age of disease onset. This trend is significant ($b=0.35\%$, $p<0.01$). $b$ is the linear regression coefficient. No significant trend was observed in men.

Table 4a HLA and seropositivity in RA: comparison of HLA frequencies in seropositive and negative patients v controls

<table>
<thead>
<tr>
<th>Seropositive patients v controls</th>
<th>Controls AF (n=108)</th>
<th>All AF (n=326)</th>
<th>RA men AF (n=107)</th>
<th>RA women AF (n=219)</th>
</tr>
</thead>
<tbody>
<tr>
<td>DR4</td>
<td>31-3</td>
<td>67-4</td>
<td>64-3</td>
<td>69-0</td>
</tr>
<tr>
<td>Dw4</td>
<td>25-3</td>
<td>51-9</td>
<td>47-3</td>
<td>54-2</td>
</tr>
<tr>
<td>DRw53</td>
<td>54-9</td>
<td>77-4</td>
<td>73-2</td>
<td>76-0</td>
</tr>
<tr>
<td>Bw62</td>
<td>8-3</td>
<td>21-5</td>
<td>28-7</td>
<td>18-0</td>
</tr>
<tr>
<td>Bw60</td>
<td>9-2</td>
<td>15-3</td>
<td>14-2</td>
<td>15-5</td>
</tr>
<tr>
<td>Cw3</td>
<td>26-0</td>
<td>33-4</td>
<td>35-1</td>
<td>32-6</td>
</tr>
<tr>
<td>A2</td>
<td>47-2</td>
<td>57-3</td>
<td>68-1</td>
<td>52-2</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Seronegative patients v controls</th>
<th>Controls AF (n=83)</th>
<th>All AF (n=326)</th>
<th>RA men AF (n=19)</th>
<th>RA women AF (n=64)</th>
</tr>
</thead>
<tbody>
<tr>
<td>DR4</td>
<td>31-3</td>
<td>54-9</td>
<td>68-7</td>
<td>50-9</td>
</tr>
<tr>
<td>Dw4</td>
<td>25-3</td>
<td>36-5</td>
<td>33-3</td>
<td>37-5</td>
</tr>
<tr>
<td>DRw53</td>
<td>54-9</td>
<td>61-9</td>
<td>75-0</td>
<td>58-1</td>
</tr>
<tr>
<td>Bw62</td>
<td>8-3</td>
<td>18-4</td>
<td>23-5</td>
<td>14-5</td>
</tr>
<tr>
<td>Bw60</td>
<td>9-2</td>
<td>26-5</td>
<td>29-4</td>
<td>25-8</td>
</tr>
<tr>
<td>Cw3</td>
<td>26-0</td>
<td>40-5</td>
<td>58-8</td>
<td>35-4</td>
</tr>
<tr>
<td>A2</td>
<td>47-2</td>
<td>63-2</td>
<td>94-1</td>
<td>54-8</td>
</tr>
</tbody>
</table>

AF=antigen frequency; $p=$probability (as in Table 2).

and 83 (20.3%) seronegative (Table 4). The frequency of DR4 was significantly raised in both seropositive and negative patients compared with controls. Dw4 and DRw53, however, were significantly raised only in the seropositive patients. The frequency of HLA-Bw60 was significantly higher in the negative patients than in controls.

A comparison of the seropositive and negative panels showed that the frequencies of DR4, Dw4, and DRw53 were significantly higher in the former group, whereas that of Bw60 was significantly higher in the latter.

The frequency of DR4 and A2 was significantly raised in both seropositive and negative male patients compared with non-RA controls. The frequencies of DRw53, Dw4, and Bw62 were also increased in seropositive men, and those of Bw60 and Cw3 in seronegative men. In the seropositive
female patients only DR4, Dw4, and DRw53 had raised frequencies and only DR4 and Bw60 were increased in the seronegative female patients.

When seropositive and negative men were compared with each other the only difference was that the frequency of A2 was significantly raised in the negative group. A comparison of seropositive and seronegative women, however, shows that the frequencies of DR4, Dw4, and DRw53 were significantly increased in the positive group, and that of Bw60 was increased in the seronegative female patients.

**HLA and Radiological Changes**

Radiological data of the hand and feet were available on 218 patients, of which 45 were non-erosive, 43 had mild erosions, 85 moderate, and 45 severe erosions; 79-36% of the total tested panel had erosions to some degree. Table 5a shows the DR4 and Dw4 frequencies in the different groups. The frequency of DR4 increases from 57-1% in the non-erosive group to 74-4% in the severe group. HLA-Dw4 increased from 40% to 66% in the same groups and that of DRw53 from 62-2% to 77-7%. The trend of increase of the Dw4 frequency with the severity of the erosions was significant (Kendall's tau=0.22, p<0.05). The trend for DR4 and DRw53 did not reach significance. The results were different in the two sexes.

A significant trend of increased frequency of each of the antigens Dw4, DR4, and DRw53 with increasing severity of erosions was seen in women only (τ=0.29, p<0.05 for Dw4, τ=0.27, p<0.01 for DR4, and τ=0.28, p<0.01 for DRw53). No significant differences were observed for any antigen in relation to the severity of erosions in the male patients.

Subluxations of the cervical spine were studied in 167 patients, of which 105 had none, and 62 had subluxation (Table 5b). The frequency of DR4 in those patients with and without subluxations was significantly different from that of the non-RA controls. The frequency of HLA-B27 was signifi-

### Table 5a  HLA and radiological changes in RA: erosions of the hands and feet

<table>
<thead>
<tr>
<th></th>
<th>None (n=45)</th>
<th>Mild (n=43)</th>
<th>Moderate (n=85)</th>
<th>Severe (n=45)</th>
<th>Tau†</th>
</tr>
</thead>
<tbody>
<tr>
<td>All</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DR4</td>
<td>57-1</td>
<td>**</td>
<td>55-0</td>
<td>65-0</td>
<td>74-4</td>
</tr>
<tr>
<td>Dw4</td>
<td>40-0</td>
<td>NS</td>
<td>33-3</td>
<td>50-0</td>
<td>66-6</td>
</tr>
<tr>
<td>DRw53</td>
<td>62-2</td>
<td>NS</td>
<td>58-1</td>
<td>74-1</td>
<td>**</td>
</tr>
<tr>
<td>A2</td>
<td>60-0</td>
<td>NS</td>
<td>44-7</td>
<td>67-1</td>
<td>**</td>
</tr>
<tr>
<td>RA men</td>
<td></td>
<td></td>
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<td>DR4</td>
<td>66-6</td>
<td>**</td>
<td>53-3</td>
<td>63-3</td>
<td>**</td>
</tr>
<tr>
<td>Dw4</td>
<td>42-8</td>
<td>NS</td>
<td>33-3</td>
<td>43-4</td>
<td>**</td>
</tr>
<tr>
<td>DRw53</td>
<td>86-6</td>
<td>**</td>
<td>60-0</td>
<td>73-3</td>
<td>**</td>
</tr>
<tr>
<td>A2</td>
<td>81-8</td>
<td>*</td>
<td>60-0</td>
<td>88-0</td>
<td>**</td>
</tr>
<tr>
<td>RA women</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DR4</td>
<td>51-8</td>
<td>*</td>
<td>56-0</td>
<td>66-0</td>
<td>**</td>
</tr>
<tr>
<td>Dw4</td>
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<td>NS</td>
<td>33-3</td>
<td>53-0</td>
<td>**</td>
</tr>
<tr>
<td>DRw53</td>
<td>55-5</td>
<td>NS</td>
<td>64-0</td>
<td>82-0</td>
<td>**</td>
</tr>
<tr>
<td>A2</td>
<td>50-0</td>
<td>NS</td>
<td>34-7</td>
<td>56-8</td>
<td>**</td>
</tr>
</tbody>
</table>

Results are expressed as phenotype frequency (%) and p value for each antigen compared with controls. p values as in Table 2. †Tau=Kendall's tau for linear trend.
HLA IN OTHER RA SUBSETS (Table 6)

Subcutaneous nodules

The frequencies of DR4, Dw4, and A2 were higher in patients with subcutaneous nodules, when compared with patients without nodules. These differences did not reach statistical significance.

Extra-articular disease

HLA-DR4, Dw4, Bw62, Cw3, and A2 were all more frequent in the patients with extra-articular manifestations other than nodules (n=35) compared with those without any extra-articular disease (n=282). The comparison between these two groups showed significant differences in the frequencies of Bw62 and Cw3, which were raised in the positive patients.

Table 6 HLA associations in other RA subsets

<table>
<thead>
<tr>
<th>Nodules</th>
<th>Controls</th>
<th>Positive v</th>
<th>Negative v</th>
<th>Positive v</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(n=108)</td>
<td>(n=108)</td>
<td>(n=287)</td>
<td></td>
</tr>
<tr>
<td>DR4</td>
<td>31-3</td>
<td>71-0***</td>
<td>64-2****</td>
<td>NS</td>
</tr>
<tr>
<td>Dw4</td>
<td>25-2</td>
<td>56-6***</td>
<td>49-0****</td>
<td>NS</td>
</tr>
<tr>
<td>DRw53</td>
<td>54-9</td>
<td>75-0**</td>
<td>71-9**</td>
<td>NS</td>
</tr>
<tr>
<td>B27</td>
<td>11-8</td>
<td>8-2 NS</td>
<td>25-4*</td>
<td></td>
</tr>
<tr>
<td>Probability (as in Table 2).</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Discussion

This analysis has confirmed positive associations between RA and HLA-DR4, Dw4, and DRw53, and negative associations with DR2 and DR7 in a large survey of British Caucasoid patients. Furthermore, a significant association exists between RA and HLA-A2, which has been tentatively suggested in early reports.6

The subdivision of RA patients into subsets according to different clinical and immunological criteria has led us and others to suggest that there is a strong relation between DR4/Dw4 and the severity of the disease as measured by the presence of rheumatoid factors,3 21-23 treatment,21 24 radiological changes,25 26 clinical status,26 27 and extra-articular manifestations.28 This is in contradiction with findings reported by other investigators.29 30 An overall association between DR4, Dw4, and DRw53 and RA severity is confirmed by our data. We have observed a significant trend between the increased severity in erosions of the hand and feet and the raised DR4/Dw4/DRw53 frequency. The trend for HLA-Dw4 is particularly clear, thus confirming and extending our previous study.25

A significantly higher frequency of DR4, Dw4, and DRw53 was found in seropositive RA patients when compared with seronegative ones. The difference in DR4 frequency was relatively small (12.5%, 98% confidence interval 7.7-17.3%). This might explain the apparently contradictory results reported from smaller series.31 32 In our analysis seropositive data from two different tests (latex and RAHA) have been pooled. Furthermore, seropositivity can be a somewhat transient feature of RA, with a proportion of false negatives being expected at any time. Both these factors may contribute to the heterogeneity of the data. In each group of data, however, seropositive patients had a consistently higher frequency of DR4/Dw4/DRw53 than that in the seronegative group. Additionally, pooling such heterogeneous data is likely to mask any differences found, and thus the real significance of our finding may be underestimated.

An interesting result in the analysis of the seronegative patients was the association found with DR4/Bw60 but not with Dw4. It is well known that this particular haplotype is associated in Caucasoids with the Dw14 specificity.33 It is therefore most likely that an association exists between this subset of the disease and the haplotype Dw14/DR4/Bw60. Although only a small number of patients were tested for the Dw14 specificity, it is interesting to note that the two Dw14 positive patients were both seronegative. Obviously these data are too limited to presume such an association, but they are an
indication that it may exist. A further study of this specificity in seronegative patients would be of interest.

Although there are no absolute diagnostic criteria for RA, a firm diagnosis becomes more likely when the disease is more severe. The more homogeneous groups of severe patients in this study showed a stronger HLA association with the disease than that observed in the early diagnosed patients. This shows the importance of patient selection for any study of HLA and RA. This could also be a reflection of a possible association of HLA with RA severity. It is therefore hard to distinguish between either an HLA link with disease susceptibility or disease severity, particularly when the patient groups studied are small or highly selected. The results of our analysis suggest that DR4 may be associated with overall RA susceptibility since an association with DR4 is seen in all subsets of patients. Furthermore, the presence of Dw4 and DRw53 in addition to DR4 appears to be associated with parameters of disease severity such as the presence of nodules and other extra-articular manifestations, erosions, and seropositivity.

The relation between the HLA status and the presence of subluxation of the cervical spine was interesting and has not previously been noted. The frequencies of DR4, Dw4, and DRw53 were not different between patients with and without subluxations. Some consider rheumatoid neck involvement occurs with the most severe disease; Rasker and Cosh, however, found that the presence of cervical subluxations was related more to the response to corticosteroid treatment than to the severity of the disease as a whole. A significant increase of the frequency of HLA-B27 in the patients with subluxation was observed. An association of B27 has been suggested in patients with 'juvenile arthritis, when the spine is involved'. Genetic 'interference' of another type of HLA related disease remains a possibility.

In addition, in most patient subsets a decrease of DR3, B8, B18, and A1 was observed. HLA-A1/B8/DR3 and B18/DR3 are common haplotypes in European Caucasoids and both are associated with a series of autoimmune disorders. A relation between RA and other autoimmune diseases is well known, often with the familial tendency for members to develop one of a range of conditions, i.e., type I diabetes, thyroiditis. It would therefore be expected that the frequency of A1, B8, B18, and DR3 would be raised in RA patients or at least have a frequency similar to that of the controls. This is not the case, however, and in fact of all the groups of patients studied the DR3 frequency was lowest in The London Hospital group (8.6% v 34.3% in controls), who were probands of RA multiplex families where a high incidence of other autoimmune disorders has been recorded. This paradox may be explained by a genetic predisposition to develop autoimmunity controlled outside the HLA region and may require the additional HLA antigens to develop a particular disorder. If this were the case, a panel of RA patients would be negatively selected for DR3 haplotypes.

There have been reports of a strong sex difference in the frequency of DR4 of Japanese RA patients, a higher DR4 frequency occurring in men than in women. We reported similar findings in a smaller group of severe RA patients. Our data now show much clearer HLA differences between male and female patients. Female patients follow the pattern seen for the overall group, i.e., high frequencies of DR4, Dw4, and DRw53, which increase in seropositive patients and in patients with erosions. In contrast, the pattern of HLA frequency distribution observed in the male patients is very different: in addition to DR4, Dw4, and DRw53, the frequencies of A2, Cw3, and Bw62 were also significantly raised. No differences between seropositive and seronegative male patients were observed, and there was no increase of the frequency of any HLA antigen in relation to the degree of erosions.

Further, the relation of HLA status and age of disease onset was different in men and women, with a decrease in DR4 frequency with increasing age of onset in women. This difference was not observed for seropositivity. No relation to the HLA status was found with the age of disease onset in the male patients.

The observations that more women than men develop RA but both show a significant increase of DR4 frequency suggest a major susceptibility factor other than HLA in female patients. This could be a hormonal factor, and it appears to be less operative in younger women. It could also be an additional genetic factor which is less operative in younger women due to some hormonal component/s. This could explain why in younger women a higher HLA association is observed, indicating the necessity of a stronger genetic component in order to develop the disease. As the woman ages this hormonal protection becomes less operative and the HLA association becomes less necessary. A possible protective factor against the development of the disease may also exist in men, where it may be overcome by a stronger genetic component (A2/Bw62/DR4). Interestingly, the age of onset of male patients does not appear to have any effect on this factor, and the association with HLA is as strong for the male patients irrespective of age of onset.
We interpret the increase of DR4/Dw4/DRw53/ Bw62/Cw3/A2 as a further confirmation of an HLA haplotype being associated with RA in men. These antigens are in linkage disequilibrium with each other in Caucasian populations. The possibility therefore exists that the high frequency of these antigens may be merely a result of the 'hitch hiking' effect due to their linkage disequilibrium with DR4. However, DR4 is also in strong linkage disequilibrium with B44 and Bw60 and with the Dw10, Dw13, and Dw14 antigens in Caucasoids. These antigens are not significantly raised in this group of male RA patients, suggesting that this particular haplotype is significantly implicated in the development of RA in men. In addition, the demonstration of a significant association between RA and HLA-A2 principally in men requires careful consideration. Although there is some linkage disequilibrium between A2 and DR4 in Caucasoids, this is weak and cannot explain the extremely high increase of the combined frequency of both A2+DR4, which is 46% in the patients compared with 18% in the controls. This A2+DR4 association is higher in men (combined antigen frequency 55%) than in female RA patients (42%). Furthermore, although the combined relative risk of A2+DR4 in the total RA panel is not higher (RR=3.37) than for DR4 alone (RR=4.0), it is higher than that for A2 alone (RR=1.6). In male RA patients the RR for A2+DR4 is 4.73, whereas the RR for DR4 alone is 3.97, and 2.8 for A2.

These data confirm that HLA is more important for the development of RA in men than in women. This difference appears to be the inferred presence of an HLA 'haplotype' DR4/Dw4/DRw53/Bw62/Cw3/A2 in Caucasoids. The 'haplotype' found in Japanese patients was Dw15/DR4/DRw53/Bw54. This appears to be due to a possible preferential allelic association of the disease with these particular antigens, rather than to linkage disequilibrium.

A multifactorial cluster analysis is now being carried out on these data in an attempt to subdivide the disease according to HLA status in addition to the clinical data.

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References


HLA and rheumatoid arthritis: a combined analysis of 440 British patients.

D Jaraquemada, W Ollier, J Awad, A Young, A Silman, I M Roitt, M Corbett, F Hay, J A Cosh and R N Maini

Ann Rheum Dis 1986 45: 627-636
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