Clinical Significance of HLA-DRB1*0410 in Japanese Patients With Idiopathic Thrombocytopenic Purpura

By Shosaku Nomura, Tatsunori Matsuzaki, Yoshih Ozaki, Manabu Yamaoka, Chie Yoshimura, Kaoruko Katsura, Gui Lan Xie, Hideo Kagawa, Tomoko Ishida, and Shirou Fukuhara

We performed HLA-A, -B, and -C antigen and -DR DNA typing in 111 Japanese patients with idiopathic thrombocytopenic purpura (ITP). DRB1*0410 was significantly increased in ITP patients compared with healthy controls (relative risk = 9.52, \(P < .05\)), but the other DRB1*04 alleles showed no significant differences. On HLA-DR serotyping, patients with Vogt-Koyanagi-Harada disease (VKH) had a high frequency of DR4, so we compared the frequencies of DRB1*04 suballeles between ITP and VKH. The high frequency of DRB1*04 was dependent on DRB1*0405 in VKH, but on DRB1*0410 in ITP. Plasma autoantibodies were studied in 111 patients using a microtiter well assay. Thirty-six patients had anti-GPIIb/IIIa autoantibodies, and antibody positivity was associated with HLA-DR4 (29 of 36, 80.6% vs 28 of 75, 37.3%) but not with DRB1*0410. When HLA-DR4 and DRB1*0410 were compared between patients with a good or poor response to prednisolone, HLA-DR4 was decreased and DRB1*0410 was significantly decreased (chi-squared = 11.455, \(P < .01\)) in patients with a good response. In conclusion, this study showed that genetically determined factors influence the course of ITP. However, our findings should be considered preliminary because of possible racial differences in HLA status between Japanese and other ITP patients.

DIOPATHIC THROMBOCYTOPENIC purpura (ITP) is a disease caused by circulating autoantibodies that react with the platelet membrane.1,2 It is thought that platelet-associated IgG is an important factor in the mechanism of ITP, since an increase of such IgG is closely related to a reduced platelet count in this disease.1,2 Although the platelet surface antigens corresponding to the antiplatelet autoantibodies involved in ITP are largely unknown, there have been several recent reports on autoantibodies to glycoprotein (GP)IIb/IIIa and GPIb,5-7 and the antigens for these GP are gradually being elucidated.8-12 Thus, the mechanism related to thrombocytopenia is becoming clear,13,14 but the etiology of ITP remains uncertain, with both genetic and environmental factors apparently involved. Based on serologic studies, associations between certain HLAs and many autoimmune diseases have long been described.15,16 Several groups have tried to establish a relationship between HLA class I or HLA-DR and chronic ITP.17-20 However, the results have been inconsistent, possibly because only a limited number of HLA-DR antigens could be determined serologically in the past.17-20

With the development of the polymerase chain reaction (PCR), identification of HLA alleles at the DNA level has become possible and has allowed more precise determination of the susceptibility epitopes showing a strong association with various autoimmune diseases.21,22 The HLA class II region encodes the heterodimeric (\(\alpha\) and \(\beta\) chains) GP expressed on the surface of antigen-presenting cells in the immune system. The T-cell receptor interacts with a complex formed by the antigenic peptide fragment bound by HLA molecules on antigen-presenting cells. The genetic polymorphism of class II molecules is known to be clustered in discrete regions of the \(\beta\) chains termed allelic hypervariable regions,23 which regulate the variability of immune responses through antigen recognition by HLA-restricted T cells. Thus, the polymorphic amino acid residues in allelic hypervariable regions of the HLA class II molecule have been suggested to have an important role in determining susceptibility or resistance to some autoimmune diseases.24,25 The HLA haplotype is also regarded as a potentially important factor in ITP,26 but its role remains unclear.

Although various methods have been used for the treatment of chronic ITP,27-31 splenectomy and administration of corticosteroids are still the mainstays of therapy.1,2 However, no parameters have been found to predict the response to treatment. In this study, we performed HLA-A, -B, and -C antigen and -DR DNA typing of Japanese ITP patients and investigated the HLA-DR4 gene variations in several clinical or pathologic subtypes of ITP. Our purpose was to determine whether anti-GPIIb/IIIa autoantibodies and the response to corticosteroid therapy are associated with specific HLA systems.

MATERIALS AND METHODS

Subjects. We studied 111 unrelated Japanese patients (29 men and 82 women) with ITP who presented to our hospital from April 1994 to December 1996. The diagnosis of ITP was made according to the standard criteria of thrombocytopenia with a normal or increased number of megakaryocytes and no evidence of any secondary cause of thrombocytopenia. The subjects were aged 22 to 78 years, and none had received a blood transfusion. Table 1 shows a brief clinical profile of the 111 ITP patients. Seventy-one controls were also randomly selected from among healthy unrelated Japanese individuals. Furthermore, 53 Japanese patients with Vogt-Koyanagi-Harada disease (VKH) were studied as disease controls. Approval was obtained from the Institutional Review Board for these studies. Informed consent was provided according to the Declaration of Helsinki.

Treatment. Prednisolone was administered first at a dose of 0.5 to 1 mg/kg daily. The response of each patient was assessed from the change in the platelet count at 6 months after starting therapy. A good response was defined as an increase in the platelet count of greater than 50 \(\times\) 10^9/L, a platelet count greater than 100 \(\times\) 10^9/L off therapy, and no more than one relapse during follow-up study. A poor response was defined as an increase in the platelet count of less than 50 \(\times\) 10^9/L. Relapse was defined as a decrease in the platelet count to less than 50 \(\times\)
HLA-DRB1*0410 AND ITP

Table 1. Clinical Characteristics of the ITP Patients

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of patients</td>
<td>111</td>
</tr>
<tr>
<td>Sex ratio (male:female)</td>
<td>29:82</td>
</tr>
<tr>
<td>Age (yr)</td>
<td>Range 22-78, Median 53</td>
</tr>
<tr>
<td>Platelet count (10^9/L)</td>
<td>3.68 ± 0.39t</td>
</tr>
<tr>
<td>PAIgG (ng/10^7)*</td>
<td>286 ± 39t</td>
</tr>
</tbody>
</table>

*PAIgG was measured by a competitive enzyme-linked immunosorbent assay (normal range, 10 to 25 ng/10^7 platelets).
†Mean ± SE.

10^7/L after a normal count had been reached. The duration of follow-up study was at least 6 months from the start of therapy.

**HLA serotyping.** ITP patients and healthy controls were subjected to serotyping for HLA class I and class II antigens using the standard complement-dependent microcytotoxicity method.32

**HLA DNA typing by PCR-restriction fragment length polymorphism.** HLA DNA typing was performed according to the manufacturer’s instructions (SMITEST HLA DNA-typing system; Sumitomo Metal, Tokyo, Japan). Genomic DNA from patients and controls was isolated by phenol extraction of sodium dodecyl sulfate–lysed and proteinase K–treated cells. DNA was amplified by the PCR procedure with Taq DNA polymerase and typed by the PCR-restriction fragment length polymorphism (RFLP) method.33 The reaction mixture was subjected to 30 cycles of denaturation for 1 minute at 96° to 97°C, annealing for 1 minute at 55° to 62°C, and extension for 2 minutes at 72°C in an automated PCR thermal sequencer (Iwaki Glass Inc, Tokyo, Japan). After amplification, aliquots of the reaction mixture were digested with allele-specific restriction endonucleases for 3 hours after addition of the appropriate reaction buffer. Samples of the cleaved and amplified DNAs were subjected to electrophoresis on 12% polyacrylamide gel in an minigel apparatus (Mupid-2; Cosmo Bio Co, Tokyo, Japan). Cleavage or noncleavage of amplified fragments was detected by staining with ethidium bromide. Discrimination of genotypes was made on the basis of RFLP band patterns thus generated.

**Detection of plasma autoantibodies against GPIIb/IIIa by microtiter well assay.** The assay was previously described by Nomura et al.34 and Kokawa et al.35 A suspension of washed platelets (1 × 10^9/µL) in 100 µg/mL leupeptin and 10 mmol/L EDTA was sonicated on ice and centrifuged at 12,500g for 30 minutes. The supernatant was solubilized in 2% Triton X-100 and centrifuged at 100,000g for 30 minutes to remove the Triton X-100-insoluble fraction. After centrifugation, the supernatant was used as the platelet lysate. Autoantibodies were assayed by a modification of the method of Woods et al.36 In brief, microtiter wells were coated with a monoclonal anti-GPIIb/IIIa antibody (NNKY1-32)36,37 by overnight incubation. The platelet lysate was then added to microtiter wells and incubated. After washing, appropriate dilutions of plasma from ITP patients (n = 111), disease controls (n = 30), or normal controls (n = 20) were added and incubated. After washing again, horseradish peroxidase–conjugated rabbit anti-human IgG was added, and the amount of IgG that bound to the platelets was determined by measuring peroxidase activity using a plate reader. Assay results were expressed in terms of the percent change in peroxidase activity above or below the level in control (normal serum) wells, using the following formula: percent change = (OD platelet extract wells − OD control wells)/OD control wells × 100.

Samples with a percent increase greater than 3 SD above the mean of 20 normal plasma samples were considered to be positive.

**Assay of platelet-associated IgG.** A competitive enzyme-linked immunosorbent assay was used to quantify platelet-associated IgG (PAIgG) in patients with ITP.38 The upper limit of normal was 25 ng/10^7 platelets.

**Statistical analysis.** The χ^2 method with continuity correction and Fisher’s exact test were used for data analysis. Relative risk was calculated according to Wolf’s method with Holdane’s correction. Briefly, it was calculated as (a × d)/(b × c), where a, b, c, and d are the number of marker-positive patients, marker-negative patients, marker-positive controls, and marker-negative controls, respectively.39

**RESULTS**

**HLA-DR serotyping.** The frequency of the HLA-DR antigens and the relative risk were calculated in 111 unrelated Japanese ITP patients and 71 unrelated Japanese controls (Table 2). DR8, DR9, and DR53 were increased in ITP patients compared with healthy controls (relative risk > 1.50). On the other hand, DR1, DR6, and DR52 were decreased in ITP patients compared with healthy controls (relative risk < 0.50). However, these differences were not statistically significant. The frequencies of DR4 and DR53 were high in both healthy controls and ITP patients. It is thought that DR53 is associated with DR4, DR7, and DR9. Because there are racial differences in HLA frequency, we studied HLA frequencies in VKH patients as Japanese disease controls. VKH is an inflammatory disease affecting multiple organs, causing bilateral panuveitis, meningoit, hearing loss, tinnitus, and vitiligo.40 It was previously reported that VKH is closely associated with DR4 and DR53 by HLA serotyping of Japanese patients.41 In the present study, DR4 and DR53 were also found in almost all VKH patients examined (94.3%).

**HLA DNA typing.** Based on the serologic data, we performed DNA typing of DR4, DR8, and DR9. HLA-DRB1 genotyping was performed by the PCR-RFLP method. DRB1*04 (DR4), *08 (DR8), and *09 (DR9) allele frequencies in ITP patients and healthy controls are shown in Table 3. DRB1*0410 was significantly increased in ITP patients compared with healthy controls (relative risk = 9.52, P < .05). However, none of the other alleles (DRB1*04, *08, and *09) showed a significant difference between ITP patients and healthy controls. HLA-DR serotyping showed that VKH patients had a high frequency of DR4, so we compared the frequency of DRB1*04 suballeles between ITP and VKH patients. Using the International Histocompatibility Workshop (1984) definitions, HLA-DR4 was classified as DR4.1 and DR4.2 subgroups with panel sera. We then compared DRB1*04 suballeles in these two subgroups among ITP patients, VKH patients, and controls.

**Table 2. HLA-DR Antigenic Frequencies of ITP Patients and Controls**

<table>
<thead>
<tr>
<th>HLA Antigen</th>
<th>Controls (n = 71)</th>
<th>ITP Patients (n = 111)</th>
<th>Relative Risk</th>
</tr>
</thead>
<tbody>
<tr>
<td>DR1</td>
<td>10</td>
<td>14.1</td>
<td>8</td>
</tr>
<tr>
<td>DR2</td>
<td>29</td>
<td>40.8</td>
<td>43</td>
</tr>
<tr>
<td>DR3</td>
<td>1</td>
<td>1.4</td>
<td>1</td>
</tr>
<tr>
<td>DR4</td>
<td>34</td>
<td>47.9</td>
<td>57</td>
</tr>
<tr>
<td>DR5</td>
<td>9</td>
<td>12.7</td>
<td>15</td>
</tr>
<tr>
<td>DR6</td>
<td>27</td>
<td>38.0</td>
<td>22</td>
</tr>
<tr>
<td>DR7</td>
<td>0</td>
<td>0.0</td>
<td>0</td>
</tr>
<tr>
<td>DR8</td>
<td>13</td>
<td>18.3</td>
<td>31</td>
</tr>
<tr>
<td>DR9</td>
<td>17</td>
<td>23.9</td>
<td>36</td>
</tr>
<tr>
<td>DR10</td>
<td>1</td>
<td>1.4</td>
<td>1</td>
</tr>
<tr>
<td>DR52</td>
<td>35</td>
<td>49.3</td>
<td>36</td>
</tr>
<tr>
<td>DR53</td>
<td>42</td>
<td>59.2</td>
<td>81</td>
</tr>
</tbody>
</table>
ITP patients showed an increase of DR1 subgroup alleles and a decrease of the DR4 subgroup. VKH patients also showed an increase of the DR4 subgroup. The high frequency of DR4 was dependent on DRB1*0405 in VKH patients, but was related to DRB1*0410 in ITP patients. Among 71 healthy controls, 34 had DRB1*04 (DR4). To analyze the suballelic distribution of DRB1*04 (DR4) more precisely in ITP and VKH patients, we investigated the DRB1*04 frequency in DR4-positive patients alone (Table 5). DRB1*0410 was again significantly increased in ITP patients compared with VKH patients and healthy controls.

Characteristics of HLA-DRB1*0410-positive ITP patients. Table 6 shows the clinical characteristics of HLA-DRB1*0410-positive ITP patients. All of them were female, and six had the homotype DRB1*0410; however, these six patients did not have any common distinguishing characteristics. The sex distribution of HLA-DRB1*0410 in our laboratory showed a slight female predominance (61.2% female), but there were no family members with HLA-DRB1*0410 and ITP. Furthermore, the frequencies of HLA-A or HLA-B antigens were not different from those in the controls. Plasma autoantibodies were studied in 111 ITP patients using a microtiter well assay, and 36 patients (32.4%) had anti-GPIIb/IIIa autoantibodies. Table 7 shows the association of HLA-DR4 with autoantibodies to GPIIb/IIIa.

There was a positive association between anti-GPIIb/IIIa antibodies and HLA-DR4 (29 of 36, 80.6% v 28 of 75, 37.3%), but there was no association with DRB1*0410. HLA-DR4 and DRB1*0410 were compared in patients with a good or poor response to prednisolone (Table 7). HLA-DR4 was slightly decreased in patients with a good response to prednisolone (poor v good, 22 of 54, 59.3% v 25 of 57, 43.9%), and DRB1*0410 was significantly decreased (poor v good, 21 of 54, 38.9% v 3 of 57, 5.3%, $\chi^2 = 11.455$, $P < .01$). However, anti-GPIIb/IIIa antibody was not correlated with the response to steroid therapy.

**DISCUSSION**

ITP is a clinically well-defined autoimmune disease caused by antiplatelet antibodies, and the antigenic epitope has recently been studied in detail, revealing that the main epitope contains GPIIb/IIIa.7-12 Various autoimmune diseases are associated with
HLA-DRB1*0410 AND ITP

Table 7. Anti-GPIIb/IIIa Autoantibody Status and Response to Prednisolone in Patients With DR4 or DRB1*0410

<table>
<thead>
<tr>
<th>Anti-GPIIb/IIIa</th>
<th>Steroid Response</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Positive (n = 36)</td>
</tr>
<tr>
<td></td>
<td>(n = 54)</td>
</tr>
<tr>
<td>DR4</td>
<td>29 80.6±</td>
</tr>
<tr>
<td>DRB1*0410</td>
<td>11 30.6±</td>
</tr>
</tbody>
</table>

*χ² = 6.954, P < .05.
†χ² = 1.862, not significant.
‡χ² = 2.098, not significant.
§χ² = 11.455, P < .01.

HLA class I and/or class II, and thus several groups have investigated the role of HLA class I and HLA-DR in ITP. However, the findings have been inconsistent, possibly because only a limited number of HLA-DR antigens could be determined serologically in the past. The polymorphisms of class II genes (HLA-DR, -DQ, and -DP) in the major histocompatibility complex can now be defined precisely by typing using the PCR-RFLP method. It was previously reported that patients with aplastic anemia who possess HLA-DR2 are more likely to respond to immunosuppressive therapy. Thus, analysis of HLA antigens and alleles may provide useful information for the treatment of autoimmune diseases. In the present study, we performed HLA typing of Japanese ITP patients to determine whether positivity for anti-GPIIb/IIIa antibodies and the response to corticosteroid therapy are associated with specific HLA systems.

We observed an increase of DR4, DR8, DR9, and DR53 in ITP patients compared with healthy controls, although these differences were not statistically significant. Since HLA frequencies show racial differences, the frequencies of DR4 and DR53 were only compared among Japanese patients. As a result, we found an increase of DR53 in both ITP and VKH patients, with a particularly marked increase in VKH. It was previously found that VKH was closely associated with DR4 and DR53 by serotyping of Japanese VKH patients. However, the association of DR4 and DR53 was not significant in our ITP patients. Furthermore, no increase of DR9 or DR7, which is known to be tightly linked to HLA-DR53, was observed in ITP patients. These results suggest that DR53 itself is unlikely to confer susceptibility to ITP.

The serologically defined HLA-DR antigen can be divided into many alleles reflecting polymorphism of the amino acid sequence in the allelic hypervariable regions of the β chain. For example, DRB1*04 can be divided into 11 alleles (DRB1*0401 to DRB1*0411). When we analyzed the frequency of HLA-DR*04 alleles, the most important finding was that DRB1*0410 was significantly increased in ITP patients compared with healthy controls (relative risk = 9.52, P < .05). Of further interest was the observation that all patients with DRB1*0410 were females. The immune response is stronger in females than in males, and there is a greater prevalence of autoimmune disease in the female population. Additionally, Cavan et al. have reported strong evidence for a sex difference in the effect of HLA markers on disease susceptibility. However, ITP is a female-predominant disease, and the sex distribution of HLA-DRB1*0410 in our laboratory showed slight female predominance (61.2% female). Thus, the relationship between DRB1*0410-positive ITP and female hormones seems to deserve further study. DRB1*04 alleles have been suggested to have an important role in determining susceptibility to several autoimmune diseases such as insulin-dependent diabetes mellitus, rheumatoid arthritis, and VKH. In particular, VKH appears to be strongly associated with DRB1*04, so we compared the frequencies of DRB1*04 alleles between our ITP and VKH patients.

We obtained the same results as in previous reports, detecting a high percentage of DRB1*0405 and DRB1*0410 (Table 4). However, DRB1*0405 was low in ITP. To confirm the significance of DRB1*0405 and DRB1*0410 in ITP and VKH patients, we compared DRB1*04 suballelic frequencies in ITP, VKH, and controls with DR4 positivity (Table 5). DRB1*0410 was also significantly increased in ITP patients compared with controls and VKH patients. The published amino acid sequences of the polymorphic β domains of DRB1 genes show that the amino acid specific for both DRB1*0405 and DRB1*0410 is the serine at position 57, instead of aspartic acid as in the other DRB1 alleles (Table 8). Amino acid sequence differences among DR4-associated alleles are confined to the COOH-terminal portion of the β domain, primarily within the three allelic hypervariable regions of the DRB1 gene. Hence, the polymorphic residues in this region are believed to be critical for both T-cell recognition and antigenic peptide binding. However, despite both DRB1*0405 and DRB1*0410 possessing the same amino acid (serine) at position 57, the frequency of DRB1*0405 was low in ITP patients compared with controls. Thus, the serine at position 57 of DRB1*0410 may not play a crucial role in the immunopathology of ITP in Japanese patients.

Next, we investigated the significance of DRB1*0410 in ITP patients. First, plasma autoantibodies were studied in 111 patients using a microtiter well assay, which showed that 36 patients (32.4%) had anti-GPIIb/IIIa autoantibodies. Many antiplatelet autoantibodies bind to platelet GPIb/IIa. In particular, there have recently been investigations on the precise localization of antigenic epitopes, such as the carboxy-terminal region (C-terminus) of GPIb. However, the Ca2+-dependent epitope of GPIb/IIa complex and the 50-kD cysteine-rich region of GPIIIa. Bowditch et al. concluded that a limited number of shared epitopes on platelet GPIb/IIa were recognized in ITP. In the present study, we observed a positive association of

Table 8. Comparison of Amino Acid Sequences in the Hypervariable Region of the DRB1 Chain

<table>
<thead>
<tr>
<th>DRB1 Residues</th>
<th>37</th>
<th>57</th>
<th>74</th>
<th>86</th>
</tr>
</thead>
<tbody>
<tr>
<td>*0403</td>
<td>Y</td>
<td>D</td>
<td>E</td>
<td>V</td>
</tr>
<tr>
<td>*0404</td>
<td>Y</td>
<td>D</td>
<td>E</td>
<td>V</td>
</tr>
<tr>
<td>*0405</td>
<td>Y</td>
<td>S</td>
<td>A</td>
<td>Gt</td>
</tr>
<tr>
<td>*0406</td>
<td>D</td>
<td>E</td>
<td>V</td>
<td></td>
</tr>
<tr>
<td>*0407</td>
<td>Y</td>
<td>D</td>
<td>E</td>
<td>G</td>
</tr>
<tr>
<td>*0410</td>
<td>Y</td>
<td>S</td>
<td>A</td>
<td>V†</td>
</tr>
</tbody>
</table>

Amino acid sequences were translated from the nucleotide sequences and are shown as the standard 1-letter codes. Dashes indicate identity with the DRB1*04 sequence.
†Different sequences in DRB1*0405 and DRB1*0410.
anti-GPIIb/IIa antibody with HLA-DR4, although we could not analyze the epitope. Thus, it seems likely that the response to the antigenic epitope of GPIIb/IIa is restricted by HLA-DR4.

Self-reactive T cells are controlled either by elimination in the thymus or by induction of tolerance in the periphery. Both mechanisms depend on the ability of antigen-presenting cells to process and present self-peptides in the context of HLA molecules, although it is now apparent that not all possible peptides from a self-antigen are presented. These limitations in processing suggest that T cells specific for some determinants on self-proteins may escape tolerance and therefore remain part of the normal T-cell repertoire. Such autoreactive T cells may only cause pathologic conditions when the presentation of normally hidden self-peptides occurs or when the immune system is confronted with foreign molecular mimics. Interestingly, autoreactive T cells to GPIIb/IIa have been isolated from ITP patients, whereas Filion et al. reported that autoreactive T cells to GPIIb/IIa are present in the peripheral blood of healthy individuals. However, there was no association of anti-GPIIb/IIa autoantibody with DRB1*0410 in the present study. There are many epitopes recognized by anti-GPIIb/IIa autoantibodies, but the clinical significance of these autoantibodies is not always clear. The lack of a correlation between anti-GPIIb/IIa antibody and the DRB1*0410 allele may suggest that this antibody does not cause ITP.

Next, we compared HLA-DR4 and DRB1*0410 distribution in patients with a good or poor response to prednisolone. HLA-DR4, particularly DRB1*0410, was significantly decreased in patients with a good response. There have been various reports on the mechanism of action of steroid hormones in ITP. Treatment with corticosteroids appears to shorten the duration of thrombocytopenia by inhibiting phagocytosis and thereby increasing the lifespan of platelets. Steroids may also directly inhibit IgG production and increase IgG catabolism. The inhibition of IgG production may result from an effect of steroids on lymphocytes, but few reports have been published on the antibodies in patients responding poorly to steroid therapy. In the present study, anti-GPIIb/IIa was not correlated with the response to steroids. A relationship between the HLA system and the outcome of therapy for ITP was previously reported by Gratama et al. Their results suggested that HLA-DR4 may predict the response to treatment, ie, a poor response to corticosteroids and a favorable outcome of splenectomy. However, they did not study DRB1*04 alleles. On the other hand, Islam et al. reported that DRB1*0405 and/or DRB1*0410 were responsible for the chronic type of VKH. The sequences of DRB1*0405 and DRB1*0410 are identical except for amino acid 86: glycine in DRB1*0405 and valine in DRB1*0410 (Table 8). Three hypervariable regions determine HLA antigenicity and possibly most of the peptide-binding capacity of HLA proteins: these regions are identical between DRB1*0405 and DRB1*0410. DR4 specificity is determined primarily by the first and second hypervariable regions (amino acids 8 to 14 and 26 to 37) of the DRB1*0410 allele. So these regions are likely to determine susceptibility to anti-GPIIb/IIa antibody in patients with ITP. Although it is difficult to conclude that amino acid 86 is the ITP-susceptibility determinant, there is a possibility that this amino acid of HLA-DRB1*0410 plays an important role in resistance to prednisolone therapy. However, other DRB1*0410-related genes may also participate in steroid resistance, so further studies of this issue are needed.

In conclusion, this was the first study to compare HLA-DR4–related alleles in pathologic and clinical subgroups of ITP. Some antigenic epitopes of GPIIb/IIa seem likely to be partially restricted by HLA-DR4, but there was no association of anti-GPIIb/IIa autoantibody with DRB1*0410. On the other hand, DRB1*0410 was significantly decreased in patients with a good response to prednisolone. Our findings indicate that genetic factors influence the clinical course of ITP, but this study should be considered preliminary because of possible racial differences in the HLA status of ITP patients from Japan and other countries.

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REFERENCES

5. Woods VL, Oh EH, Mason D, McMillan R: Autoantibodies against the platelet glycoprotein Iib/IIa complex in patients with chronic ITP. Blood 63:368, 1984
42. Petersdorf EW, Smith AG, Mickelson EM, Martin PJ, Hansen JA: Ten HLA-DR alleles defined by sequence polymorphisms within the DRB1 first domain. Immunogenetics 33:267, 1991
49. Semple JW, Freedman J: Increased antiplatelet T helper lympho-
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...cyte reactivity in patients with autoimmune thrombocytopenia. Blood 78:2619, 1991


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