HLA-DP and IL28B Polymorphisms: Influence of Host Genome on Hepatitis B Surface Antigen Seroclearance in Chronic Hepatitis B

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**Background.** The roles of single-nucleotide polymorphisms (SNPs) at HLA-DP and IL28B loci on hepatitis B surface antigen (HBsAg) seroclearance in chronic hepatitis B (CHB) infection are unknown.

**Methods.** We compared the HLA-DP (rs3077, rs9277378, rs3128917) and IL28B (rs12979860, rs8099917) polymorphisms of 203 CHB patients achieving spontaneous HBsAg seroclearance with 203 age- and sex-matched CHB patients without HBsAg seroclearance (controls).

**Results.** The distribution of all 5 polymorphisms was in Hardy-Weinberg equilibrium. HLA-DP rs3077 was associated with HBsAg seroclearance in terms of allelic frequency (minor allele A vs major allele G, \( P = .035 \); odds ratio [OR], 0.699; 95% confidence interval [CI], .501–.976) and genotypic frequency (AA vs GG/GA, \( P = .014 \); OR, 0.295; 95% CI, .106–.822). Haplotype analysis of HLA-DP polymorphisms showed haplotype block GAT (rs3077/rs9277378/rs3128917) to be associated with HBsAg seroclearance (OR, 2.17; 95% CI, 1.06–4.45, \( P = .034 \)). Influence of HLA-DP polymorphisms on HBsAg seroclearance was more pronounced in younger patients, with the OR for rs3077 minor allele A and haplotype block GAT being 0.560 and 2.68, respectively, among patients aged <50 years (\( P = .027 \) and \( P = .047 \), respectively). IL28B haplotype block CG (rs12979860/rs8099917) was associated with HBsAg seroclearance (OR, 10.5, \( P = .026 \)). None of the 5 polymorphisms influenced anti-HBs positivity among patients achieving HBsAg seroclearance, or serum HBV DNA and HBsAg titers among controls (\( P > .05 \)).

**Conclusions.** Specific SNPs in HLA-DP and IL28B locus, through individual and haplotype analysis, were associated with a higher chance of HBsAg seroclearance in CHB infection. The associations were more prominent in patients with HBsAg seroclearance at a younger age.

**Keywords.** HBsAg seroclearance; HBV; HLA; IL28B; SNP.

Seroclearance of hepatitis B surface antigen (HBsAg) is an uncommon event in the natural history of chronic hepatitis B (CHB), with its incidence ranging from 0.62% to 2.26% per year [1–3]. It is generally associated with a quiescent disease course and favorable prognosis [4], and with a lower risk of hepatocellular carcinoma if HBsAg seroclearance occurs early [5]. Although satisfactory disease control is achievable with current therapeutic options for CHB, HBsAg seroclearance remains an uncommon event during pegylated interferon therapy [6, 7] and is even more rarely seen during nucleoside analogue therapy [8, 9].

Recent natural history studies have attempted to identify clinical parameters predictive of spontaneous HBsAg seroclearance. These include a low baseline serum HBsAg level, an increased rate of serum HBsAg reduction, and a persistently undetectable viral load [2, 3, 10]. Nevertheless, these serologic and virologic parameters are only surrogate markers of sustained immune control in CHB, as the difference in magnitude of...
immune control among CHB patients is most probably related to different constituents of human genomic profiles. Studies aiming to identify host genetic determinants of HBsAg seroclearance are thus warranted.

Certain human leukocyte antigen (HLA) phenotypes have been previously characterized with persistence of hepatitis B virus (HBV) infection [11], and the recent application of genome-wide association studies has identified single-nucleotide polymorphisms (SNPs) in the HLA-DP loci to be associated with persistence of HBV infection [12]. This association is subsequently verified in the Han Chinese population comparing HBsAg positive subjects with HBsAg negative−, antibody to hepatitis B core antigen (anti-HBc) or antibody to HBsAg (anti-HBs)−positive individuals [13]. Other SNPs of interest are IL28B polymorphisms, which are known to influence outcomes in chronic hepatitis C virus (HCV) infection, including its spontaneous clearance [14] and treatment response [15]. Hepatic interferon-stimulated gene (ISG) expression is strongly associated with IL28B polymorphisms in HCV infection [16], and may also play a role in the successful immune control of HBV [17]. There is recent evidence indicating that IL28B polymorphisms may predict hepatitis B e antigen (HBeAg) seroconversion [18] and HBsAg seroclearance [19] in CHB during pegylated interferon therapy. Whether or not this is applicable to spontaneous HBsAg seroclearance has not been established.

We hence designed a case-control study including a large number of CHB patients with HBsAg seroclearance to determine whether HLA-DP and IL28B polymorphisms could influence HBsAg seroclearance in CHB.

**METHODS**

**Patients**

In the present study, we recruited CHB patients with spontaneous HBsAg seroclearance, as well as age- and sex-matched CHB patients without HBsAg seroclearance as controls. The composition of the current population has been described previously in a study on the predictive value of serum HBsAg titers with respect to subsequent HBsAg seroclearance [10]. In brief, both groups of patients were followed up at the Liver Clinic, Department of Medicine, the University of Hong Kong, Queen Mary Hospital regularly for at least 3 years. All patients were of Chinese ethnicity, had HBsAg positivity documented for >6 months, and were all HBeAg-negative on presentation to our clinic. HBsAg seroclearance was observed in the first group of patients between June 2001 and February 2011; these patients were then followed up regularly until June 2012 for their latest liver biochemistry and serum anti-HBs status. The control group, recruited between May 2010 and May 2011, was age- and sex-matched with the patient group achieving HBsAg seroclearance. No treatment had been given for both groups of patients. Patients with concomitant liver diseases and human immunodeficiency virus infection were excluded.

HBsAg seroclearance was defined as loss of serum HBsAg with or without appearance of anti-HBs for 2 samples taken at least 6 months apart. All patients consented to collection and storage of serum, with serum samples collected at each visit stored at −20°C until tested.

This study was approved by the Institutional Review Board, the University of Hong Kong and West Cluster of Hospital Authority, Hong Kong.

**HLA-DP and IL28B Polymorphisms**

The 2 polymorphisms that were first documented to be associated with chronicity of HBV infection are rs3077 and rs9277535, located in the HLA-DPA1 and HLA-DPB1 regions, respectively, of chromosome 6 [12]. SNP rs3077 was thus selected for our study. Owing to the difficulty of DNA extraction from stored sera for SNP rs9277535, we instead chose a polymorphism rs9277378, also located in HLA-DPB1, which has a high level of linkage disequilibrium (D' = 1.00, R2 = 0.954) with rs9277535 based on Haploview software version 4.2 [20] (Supplementary Figure 1). We included a third SNP rs3128917 in the HLA-DPB1 region, as this polymorphism had the highest odds ratio (OR) among 11 SNPs noted to influence chronicity of HBV infection in one study involving Han Chinese CHB patients [13]. Concerning IL28B polymorphisms, we selected SNPs rs12979860 and rs8099917, both located near the IL28B gene in chromosome 19. Both SNPs were noted to affect response to interferon-based therapy in chronic hepatitis C [15, 21]. Moreover, SNP rs12979860 was noted to influence outcomes during pegylated interferon therapy in CHB [18, 19].

All 5 SNPs were genotyped using TaqMan SNP genotyping assay (Life Technologies, Carlsbad, California). In brief, Purelink Genomic DNA Mini Kit (Life Technologies) was used to extract free circulating DNA from 200 μL of serum samples, with its concentration measured by NanoDrop 2000c spectrophotometer (Thermo Scientific, Wilmington, Delaware). SNP genotyping was performed using 5 ng of template DNA and the Quantifast Probe PCR Kit (Qiagen GmbH, Hilden, Germany), together with SNP-specific primers and FAM- and VIC-labeled probes, followed by real-time polymerase chain reaction and SNP analysis using RotorGene Q PCR System (Qiagen).

The possible genotypes for each biallelic polymorphism are as follows: rs3077 and rs9277378: GG, GA, AA (minor allele = A); rs3128917: GG, GT, TT (minor allele = T); rs12979860: CC, CT, TT (minor allele = T); and rs8099917: TT, TG, GG (minor allele = G).

**Laboratory Assays**

Serum HBV DNA levels were measured using Cobas TaqMan assay (Roche Diagnostics, Branchburg, New Jersey), with a
lower limit of detection of 20 IU/mL. Serologic markers, including serum HBeAg, anti-HBs and antibody to HBeAg (anti-HBe) were measured by Abbott Laboratories (Chicago, Illinois). The lower limit of detection for anti-HBs was 10 mIU/mL. Serum HBsAg levels were measured using the Elecsys HBsAg II assay (Roche Diagnostics, GmbH, Mannheim, Germany) with a lower limit of detection of 0.05 IU/mL.

**Statistical Analyses**

All continuous variables were expressed in median (range). For patients with undetectable serum HBV DNA or HBsAg, the results were taken as the lower limit of detection (1.30 log IU/mL and −1.30 log IU/mL, respectively). The Mann-Whitney U test, or the Kruskal-Wallis H test when appropriate, was used for continuous variables with a skewed distribution. The Pearson χ² test, or Fisher exact test when appropriate, was used for categorical variables. Odds ratios were calculated using the Cochran-Mantel-Haenszel test.

Hardy-Weinberg equilibrium for the genotypic distribution of both HLA-DP and IL28B polymorphisms in both patient groups was determined. When comparing the 2 patient groups, the following association analytical methods were used: comparing allelic frequencies (major allele “A” vs minor allele “B”), genotypic frequencies (AA vs AB vs BB), dominant gene action (AA vs AB + BB), and recessive gene action (AA + AB vs BB).

Haplotype analysis was performed by analyzing the 3 HLA-DP polymorphisms and the 2 IL28B polymorphisms together as haplotype blocks. Compared to single SNP association analysis, haplotype-based association analysis are more sensitive [22], and could capture additional phenotype-related variants, especially in loci with high linkage disequilibrium like the HLA region [23]. Haplotype frequencies were estimated with the expectation-maximization algorithm, with omnibus tests for haplotype association and haplotype-specific ORs calculated by haplotype replacement regression. If certain polymorphisms were found to be significant in individual association analyses, these polymorphisms would be controlled to determine the independent haplotype effect. Interaction between the HLA-DP and IL28B loci was examined for allelic epistasis using logistic regression.

All statistical analyses were performed using SPSS version 19.0 (SPSS Inc, Chicago, Illinois) and PLINK software version 1.07 [24]. A 2-sided P value <.05 was considered statistically significant.

**RESULTS**

**Patients**

The clinical characteristics of the 2 patient groups (n = 203 each) are depicted in Table 1. Patients achieving HBsAg seroclearance, when compared to the control group, had significantly higher HBV DNA and HBsAg levels (P < .001). There were no significant differences in the distribution of age, sex, and liver biochemistry among the 2 patient groups (all P > .05). In the group with HBsAg seroclearance, the median duration from presentation at our clinic to HBsAg seroclearance was 81.3 months (range, 36.6 to 315.3 month). The median age of HBsAg seroclearance was 51.9 years. Only 1.9% (4 of 203) patients developed anti-HBs positivity at the date of HBsAg seroclearance, although subsequently 50.2% (102 of 203) developed anti-HBs after a median follow-up period of 74.9 months (range, 11.6 to 154.7 months).

**Table 1. Clinical Demographics of the 2 Patient Groups**

<table>
<thead>
<tr>
<th>Demographic</th>
<th>Patients With HBsAg Seroclearance</th>
<th>Controls</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of patients</td>
<td>203</td>
<td>203</td>
<td>. . .</td>
</tr>
<tr>
<td>Age, y</td>
<td>51.9 (16.6–82.4)</td>
<td>52.2 (22.7–80.4)</td>
<td>.784</td>
</tr>
<tr>
<td>No. of male patients (%)</td>
<td>143 (70.4)</td>
<td>143 (70.4)</td>
<td>1.000</td>
</tr>
<tr>
<td>Albumin, g/dL</td>
<td>45 (36–53)</td>
<td>44 (37–50)</td>
<td>.157</td>
</tr>
<tr>
<td>Bilirubin, μmol/L</td>
<td>11 (4–73)</td>
<td>10 (2–50)</td>
<td>.309</td>
</tr>
<tr>
<td>ALT, U/L</td>
<td>22 (10–77)</td>
<td>26 (9–97)</td>
<td>.176</td>
</tr>
<tr>
<td>HBsAg, log IU/mL</td>
<td>−1.30</td>
<td>2.60 (−1.17 to 4.35)</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>HBV DNA, log IU/mL</td>
<td>1.30 (1.30–2.80)</td>
<td>3.25 (1.30–8.29)</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>No. of patients with undetectable HBV DNA (%)</td>
<td>175 (86.2)</td>
<td>19 (9.4)</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>Number of patients with detectable anti-HBs (%)</td>
<td>102 (50.2)b</td>
<td>. . .</td>
<td>. . .</td>
</tr>
</tbody>
</table>

Continuous variables expressed as median (range).

Abbreviations: ALT, alanine aminotransferase; anti-HBs, antibody to the hepatitis B surface antigen; HBsAg, hepatitis B surface antigen; HBV, hepatitis B virus.

Lower limit of detection: HBsAg 0.05 IU/mL (or −1.30 log IU/mL), HBV DNA 20 IU/mL (or 1.30 log IU/mL).

a The youngest available control recruited during enrollment period (22.7 years) was matched with the youngest patient achieving HBsAg seroclearance (16.6 years).

b Detected after a median follow-up time of 74.9 months (range, 11.6 to 154.7 months) after HBsAg seroclearance.
The genotypic distribution of all 5 polymorphisms is depicted in Figure 1. The minor allelic frequencies (MAFs) of each SNP were as follows: rs3077, 0.223; rs9277378, 0.239; rs3128917, 0.218; rs12979860, 0.070; rs8099917, 0.063. The Hardy-Weinberg calculations for all 5 polymorphisms are shown in Supplementary Table 1. All 5 polymorphisms in both patient groups were in Hardy-Weinberg equilibrium (all $P > .05$).

### Association Analysis of HLA-DP and IL28B Polymorphisms With HBsAg Seroclearance

The associations of the 5 individual SNP polymorphisms with HBsAg seroclearance are shown in Table 2. HLA-DP rs3077 was associated with HBsAg seroclearance, with minor allele “A” associated with a reduced probability of HBsAg seroclearance (allelic frequency model: $P = .035$; OR, 0.699; 95% confidence interval [CI], .501–.976; recessive gene model: $P = .013$; OR, 0.528; 95% CI, 0.315–0.822). The remaining 2 HLA-DP polymorphisms and both IL28B polymorphisms had no association with HBsAg seroclearance (all $P > .05$).

The ORs of HLA-DP rs3077 minor allele “A” with HBsAg seroclearance within different age ranges is depicted in Figure 2A. The probability of HBsAg seroclearance was further reduced with younger age; among patients aged <50 years, the OR of HBsAg seroclearance was 0.560 (95% CI, .333–.940; $P = .027$). HLA-DP rs3077 had no association with HBsAg seroclearance among patients aged ≥50 years (allelic frequency model $P = .617$).

There was no significant association between HLA-DP rs3077 and HBsAg seroclearance if solely male (n = 286) and female (n = 120) patients were analyzed ($P = .091$ and $P = .190$ respectively for allelic frequency model).

### Haplotype Analysis of HLA-DP and IL28B Polymorphisms With HBsAg Seroclearance

Haplotypes were constructed based on the 3 HLA-DP and 2 IL28B polymorphisms and were analyzed for their associations with HBsAg seroclearance, with the results depicted in Table 3. Based on the HLA-DP haplotype constructed from SNPs rs3077 (minor allele “A”), rs9277378 (minor allele “A”), and...
rs3128917 (minor allele “T”) (Table 3A), when comparing the target haplotype with remaining haplotypic combinations, haplotype block GAT was significantly associated with HBsAg seroclearance (P = .034; OR, 2.17; 95% CI, 1.06–4.45). After controlling SNP rs3077, there was still a trend toward significance (P = .060). All other haplotypic combinations showed no significance with HBsAg seroclearance.

The ORs of haplotype block GAT with HBsAg seroclearance among different age ranges is shown in Figure 2B. An increasing OR was seen among patients of younger age. Among patients aged <50 years, haplotype block GAT had an OR of 2.68 (95% CI, 1.01–7.13, P = .047). Haplotype GAT had no association with HBsAg seroclearance among patients aged 50 years or older (P = .394), male patients (P = .115), and female patients (P = .141).

Analysis of the IL28B haplotype constructed from SNPs rs12979860 (minor allele “T”) and rs8099917 (minor allele “G”) and its association with HBsAg seroclearance is depicted in Table 3B. When comparing the target haplotype with remaining haplotypic combinations, haplotype block CG was significantly associated with HBsAg seroclearance (P = .026; OR, 10.5; 95% CI, 1.33–82.5). Only 1 individual from the control group (0.23%), a woman aged 50.5 years, had the IL28B CG haplotype.

Interaction Analysis Between HLA-DP and IL28B Polymorphisms

Epistasis testing between HLA-DP and IL28B locus is depicted in Table 4. Significant interaction was demonstrated only between HLA-DP rs9277378 and IL28B rs8099917 (OR, 3.57, P = .031). Other combinations between the 2 loci did not reveal significant interaction (P > .05).

Our previous haplotype analysis suggested HLA-DP rs9277378 minor allele “A” and IL28B rs8099917 minor allele “G” were associated with HBsAg seroclearance. Among patients with HBsAg seroclearance, 8.37% had both minor alleles present, compared to 3.94% in controls, with a trend toward significance (P = .063).

Relationship Between HLA-DP and IL28B Polymorphisms With Serum Anti-HBs, HBV DNA, and HBsAg Levels

In patients achieving HBsAg seroclearance, none of the 5 polymorphisms had any association with subsequent anti-HBs positivity (Supplementary Table 2A, all P > .05). Minor allele “T” of HLA-DP polymorphism rs3128917 was significantly associated with a reduced probability of detectable HBV DNA at HBsAg seroclearance (P = .042; OR, 0.432; 95% CI, .189–.989). The remaining 4 polymorphisms did not show any association with serum HBV DNA detectability among patients achieving...
HBsAg seroclearance (all \( P > .05 \)). In the control group (Supplementary Table 28), there was no significant difference in median HBV DNA levels and HBsAg levels when comparing the allelic and genotypic distribution of all 5 polymorphisms (all \( P > .05 \)).

**DISCUSSION**

In this case-control study consisting of a large population of CHB patients with spontaneous HBsAg seroclearance and matched controls, we demonstrated that HLA-DP and IL28B
polymorphisms were associated with HBsAg seroclearance in CHB, illustrating the importance of host factors in influencing HBsAg seroclearance. Previous studies investigated genetic variants and HBV clearance from acute HBV infection [13, 25, 26]. To our knowledge, the present study is the first large population study to examine the roles of host genetic variants on spontaneous HBsAg seroclearance in chronic hepatitis B in whom HBsAg seroclearance is a rare event. The matching of age in our 2 patient groups meant infection duration was controlled, as Asian CHB patients acquire the disease at a very young age [27].

Our study findings showed the minor allele “A” of HLA-DP rs3077 to be associated with a reduced probability of HBsAg seroclearance (OR, 0.699; P = .035). In addition, haplotype analysis of the 3 studied HLA-DP polymorphisms also demonstrated the haplotype block GAT to be associated with HBsAg seroclearance (OR, 2.17; P = .034). Due to the persistence of intrahepatic covalently closed circular HBV DNA (cccDNA) [5], HBsAg seroclearance rates in chronic infection (0.62%–2.26% per year) are much lower than in acute HBV infection (>90% in adults) [28]. We believe favorable HLA-DP polymorphisms are able to strengthen the host immune response to counteract the viral persistence of cccDNA. Similar to other classical HLA class II molecules [29], HLA-DPs promote antigen presentation to CD4-positive helper T cells by promoting T-cell allore cognition and peptide binding [30], resulting in an enhanced immune response and HBV clearance. Although both CD4+ helper T cells and CD8+ cytotoxic T cells are the master regulators of the adaptive immune response to HBV, with the role of CD8+ T cells in mediating HBV clearance dependent on the presence of CD4+ T cells [31].

The involvement of HLA-DP polymorphisms is not only evident at the level of cellular function, but also at the level of gene expression. HLA-DP polymorphisms influence the level of messenger RNA (mRNA) expression of HLA-DPA1 and HLA-DPB1 molecules in the liver [32]. SNP variants are able to regulate the stability and splicing of mRNAs [33], with a recent study showing a variant in the 3’ untranslated region of HLA-DPB1 mRNA associated with HBV clearance [34]. The exact mechanistic nature of HLA-DP polymorphisms and HBV clearance would need to be addressed in future functional and expressional studies.

The positive association of IL28B haplotype CG with HBsAg seroclearance (Table 3B) is an intriguing finding. IL28B is a member of the interferon-λ family, and interferon-λ is involved in the upregulation of ISGs [35], which could lead to the inhibition of HBV [36]. Therefore, similar to the scenario of interferon therapy [19], IL28B could also play a role in spontaneous HBsAg seroclearance. This is further supported by the presence of interaction found between IL28B and HLA-DP (Table 4). Nevertheless, the MAFs of IL28B polymorphisms in subjects of Chinese ethnicity are <10% [37], resulting in an increased error margin as demonstrated by the wide confidence intervals obtained. Association studies involving CHB patients of European descent, in whom IL28B MAFs are higher, would better define the predictability of IL28B polymorphisms in HBsAg seroclearance.

Genotyping HLA-DP polymorphisms could have an impact on the clinical management of CHB. With a higher probability of HBsAg seroclearance, patients with favorable HLA-DP polymorphisms (and similarly for the IL28B CG haplotype) could be potential candidates for pegylated interferon therapy. This is especially significant among patients aged <50 years, in which

### Table 3. Haplotype Analysis of the 3 HLA-DP Polymorphisms^{ab}

<table>
<thead>
<tr>
<th>Haplotype</th>
<th>Frequency (Case/ Controls)</th>
<th>Odds Ratio</th>
<th>95% CI</th>
<th>P Value</th>
<th>rs3077 Controlled PValue</th>
</tr>
</thead>
<tbody>
<tr>
<td>GGG</td>
<td>0.622/0.599</td>
<td>1.13</td>
<td>.825–1.48</td>
<td>.503</td>
<td>.268</td>
</tr>
<tr>
<td>GAG</td>
<td>0.047/0.040</td>
<td>1.25</td>
<td>.560–2.62</td>
<td>.625</td>
<td>.828</td>
</tr>
<tr>
<td>GAT</td>
<td>0.069/0.037</td>
<td>2.17</td>
<td>1.06–4.45</td>
<td>.034</td>
<td>.060</td>
</tr>
<tr>
<td>GGT</td>
<td>0.072/0.071</td>
<td>1.07</td>
<td>.584–1.86</td>
<td>.890</td>
<td>.834</td>
</tr>
<tr>
<td>AGG</td>
<td>0.064/0.094</td>
<td>0.655</td>
<td>.384–1.12</td>
<td>.120</td>
<td>.736</td>
</tr>
<tr>
<td>AAG</td>
<td>0.036/0.062</td>
<td>0.478</td>
<td>.226–1.01</td>
<td>.054</td>
<td>.237</td>
</tr>
<tr>
<td>AAT</td>
<td>0.090/0.097</td>
<td>0.926</td>
<td>.564–1.52</td>
<td>.761</td>
<td>.208</td>
</tr>
<tr>
<td>CG</td>
<td>0.899/0.934</td>
<td>0.634</td>
<td>.382–1.05</td>
<td>.079</td>
<td></td>
</tr>
<tr>
<td>GT</td>
<td>0.025/0.002</td>
<td>10.5</td>
<td>1.33–82.5</td>
<td>.026</td>
<td></td>
</tr>
<tr>
<td>TT</td>
<td>0.032/0.010</td>
<td>2.79</td>
<td>.950–8.18</td>
<td>.062</td>
<td></td>
</tr>
<tr>
<td>TG</td>
<td>0.044/0.054</td>
<td>0.81</td>
<td>.427–1.53</td>
<td>.518</td>
<td></td>
</tr>
</tbody>
</table>

(A) Order of haplotype block: rs3077 (minor allele “A”), rs9277378 (minor allele “A”), rs3128917 (minor allele “A”). Odds ratios compared target haplotype vs the remaining haplotypic combinations.

(B) Order of haplotype block: rs12979860 (minor allele “T”), rs8099917 (minor allele “G”). Odds ratios compared target haplotype vs the remaining haplotypic combinations.

Abbreviation: CI, confidence interval.

^{a} The 2 IL28B polymorphisms.

^{b} With hepatitis B surface antigen seroclearance.

### Table 4. Epistasis Testing Between HLA-DP and IL28B Polymorphisms

<table>
<thead>
<tr>
<th>HLA-DP SNP</th>
<th>IL28B SNP</th>
<th>OR</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>rs3077</td>
<td>rs12979860</td>
<td>0.959</td>
<td>.919</td>
</tr>
<tr>
<td>rs3077</td>
<td>rs8099917</td>
<td>0.951</td>
<td>.925</td>
</tr>
<tr>
<td>rs9277378</td>
<td>rs12979860</td>
<td>2.54</td>
<td>.070</td>
</tr>
<tr>
<td>rs9277378</td>
<td>rs8099917</td>
<td>3.57</td>
<td>.031</td>
</tr>
<tr>
<td>rs3128917</td>
<td>rs12979860</td>
<td>2.44</td>
<td>.117</td>
</tr>
<tr>
<td>rs3128917</td>
<td>rs8099917</td>
<td>3.08</td>
<td>.081</td>
</tr>
</tbody>
</table>

Abbreviations: OR, odds ratio; SNP, single-nucleotide polymorphism.
the probability of favorable HLA-DP polymorphisms being associated with HBsAg seroclearance is particularly pronounced (Figures 2A and 2B). Patients with favorable HLA-DP polymorphisms and on nucleoside analogue therapy could be foretold HBsAg seroclearance is a potentially achievable target and thus lifelong treatment might not be required when the ultimate treatment endpoint of HBsAg loss is achieved [38].

This study is limited by the lack of data on liver histology and HBV genotyping. Nevertheless, our Chinese patient cohort (based on 100 matched patients in each arm) [10] and other studies [3] have demonstrated that HBV genotyping does not affect HBsAg seroclearance in the Chinese CHB population. Recruiting CHB patients from different ethnic populations would be more important in future studies, as the degree of genetic variations differs among individuals of different ethnicities [39]. Although our study did not find any association between genetic variants and anti-HBs positivity, HBV DNA levels or HBsAg levels, these results should also be validated longitudinally in the future, especially as the development of anti-HBs may be related to the duration of follow-up.

In conclusion, host genome variants influence spontaneous HBsAg seroclearance in CHB, as illustrated by individual analysis involving HLA-DP rs3077 and haplotype analysis of both HLA-DP and IL28B. The effect of HLA-DP polymorphisms is increased among patients with younger age (especially <50 years). Further studies aimed at sequencing loci adjacent to the candidate genes identified in this study may reveal additional genetic determinants that influence a favorable disease outcome in CHB.

Supplementary Data

Supplementary materials are available at Clinical Infectious Diseases online (http://cid.oxfordjournals.org/). Supplementary materials consist of data provided by the author that are published to benefit the reader. The posted materials are not copyrighted. The contents of all supplementary data are the sole responsibility of the authors. Questions or messages regarding errors should be addressed to the author.

Notes

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Potential conflicts of interest. All authors: No reported conflicts.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

References


