Differential Distribution of HLA Alleles in Two Forms of Guillain-Barré Syndrome

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Guillain-Barré syndrome in northern China occurs in two forms: acute inflammatory demyelinating polyneuropathy (AIDP) and acute motor axonal neuropathy (AMAN). AMAN and AIDP have an immunologic basis, and some cases are associated with preceding *Campylobacter jejuni* infection. The distribution of allelic forms of the histocompatibility genes HLA-DPB1, DQB1, DRB1, DRB3, DRB4, and DRB5 was examined by DNA-based technology in 34 control, 12 AIDP, and 31 AMAN cases. In AIDP patients, the DRB1∗1301 allele showed a significant increase (18% vs. 0%, $P = .055$). In AMAN patients, alleles DRB1∗1301–03 and DRB1∗1312, taken collectively, were increased (19% vs. 0%, $P = .009$), but by itself, the DRB1∗1301 allele was not increased, as in AIDP patients. With a larger number of persons, more definitive statements will be possible; however, the differential distribution of DR13 allelic forms between AIDP and AMAN cases may suggest that there are different immunologic mechanisms operating at the molecular level of these diseases.

Guillain-Barré syndrome (GBS) has been considered to be primarily an acute inflammatory demyelinating polyneuropathy (AIDP). In North America and Europe, the disease is sporadic and nonseasonal and affects persons of all ages [1]; however, reports from China describe patients whose clinical characteristics resemble those of patients with demyelinating GBS and whose epidemiologic features differ from those of patients seen in North America and Europe [2–5]. These Chinese cases occur largely among rural residents of northern China, predominantly during the summer and primarily in children.

Recent studies, using electrodiagnostic criteria, suggest that GBS in China occurs in two forms, AIDP and acute motor axonal neuropathy (AMAN) [6, 7]. Both forms have relatively good prognoses, with similar time courses of improvement [8]. The disease is of unknown etiology, although several lines of evidence suggest an immunologic pathogenesis. Approximately two-thirds of the cases are preceded by an infection, most frequently of the respiratory or gastrointestinal tracts. Characteristically, 76% of the AMAN and 42% of the AIDP GBS patients have serologic evidence consistent with recent *Campylobacter jejuni* infection [8]. In addition, significantly more GBS patients than village controls have anti-glycolipid antibodies (IgG anti-GM_1; 42% vs. 6%, $P < .01$) [8]. No clear relationship has been established between GBS, antecedent *C. jejuni* infection, and anti-GM_1 antibodies [9, 10]. Furthermore, in AIDP cases, lymphocytic infiltration and macrophage-mediated demyelination have been observed, while in AMAN cases, a prominent feature has been the presence of macrophages within the periaxonal space, surrounding or displacing the axon and surrounded by an intact myelin sheath [7].

These differences in pathologic findings between AIDP and the axonal patterns are likely to reflect differences in the pathogenetic mechanisms. Of interest, those who develop GBS are only a minority of those who become infected by different microbial agents. It has been hypothesized that this might be the case because there may be disease-susceptibility genes that are responsible for predisposing certain individuals, while other persons with the normal equivalent to these genes recover without developing postinfectious polyneuropathy. A candidate group of genes that are very polymorphic and intimately involved in the immune response are the genes that code for histocompatibility (HLA) molecules.

HLA antigens are glycoproteins coded by genes within the human major histocompatibility complex, which is on the short arm of chromosome 6 and includes both class I and class II molecules. These antigens are expressed on the surface of cells involved in the presentation of peptides to the T cell receptors of either $CD8^+$ cells (through class I molecules) or $CD4^+$ T cells (through class II molecules). Recognition of the HLA-peptide complex by the T cell receptor initiates a cascade of

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reactions known as ''the antigen-specific immune response.'' Because of their central role in the immune response, these molecules are associated with disease susceptibility to a greater extent than is any other known genetic marker in humans. The hallmark of these molecules is their polymorphism, and different diseases seem to be associated with different alleles. The recently developed technology of defining polymorphism by DNA-based techniques has improved our capability for determining these alleles and for identifying many more that previously were not detectable. This technology has also contributed to the identification of specific HLA alleles and specific amino acids within the binding cleft of the HLA molecules that may be associated with diseases. The technology has allowed for the identification of specific amino acid positions on the HLA molecules as possible susceptibility factors in insulin-dependent diabetes [11, 12], rheumatoid arthritis [13], tuberculoid leprosy [14], and berylliosis [15].

In the past, there have been a limited number of studies on the association of GBS with HLA molecules [16–19]. Only one study from Mexico suggests a possible association of GBS with DR3 [20]. A recent study [21] revealed an association between HLA-DQB1∗03 and GBS with preceding *C. jejuni* infection. The same association was not present among patients who were seronegative for *C. jejuni.* However, none of these previously reported studies have evaluated allele frequencies in the separate forms of GBS, specifically AIDP and AMAN. Given the improved technology for identifying HLA alleles and given that we can distinguish these two seemingly similar neuropathies (GBS from AMAN) by other criteria, it is meaningful to evaluate whether there is any particular HLA association with each of these diseases.

Methods

Forty-three patients with GBS were recruited from northern China and carefully characterized [6, 7]. Electrodiagnostic studies showed that 12 had AIDP and 31 had AMAN. Thirty-four control subjects from the same area were also included in the study. Blood from all these subjects was available on a filter paper (Whatman #1). Blood was removed from the filter, and DNA was prepared as previously described [22]. High-resolution DNA-based HLA typing was done to identify DRB1, DRB3, DRB4, DRB5, DQB1, and DPB1 alleles [22].

The χ^2 test with Yates's correction was used to evaluate the differences of the allelic frequencies between various groups. To correct for the multiple comparisons carried out, the apparent significance level was multiplied by the number of statistical tests [23]. Fisher's two-tailed exact test was used to determine the statistical significance of the differences when the numbers of subjects involved were very low. When the frequency was 0 in a particular group, the approximation of Haldane [24, 25] was used.

Results

As shown in table 1, allele DRB1∗1301 was found in 18% of patients with AIDP and in 0% of controls ($P = .055$), and alleles DRB∗1301-1303 and DRB1∗1312, collectively, were found in 19% of patients with AMAN and in 0% of controls ($P = .009$). In addition, there was a differential distribution of the DQB1∗06 allele among AIDP (25%), AMAN (55%), and control (27%) cases. Statistical analysis indicated that the frequency of DQB1∗06 among control and AMAN cases differed significantly ($P = .018$). However, this difference lost its strength after it was multiplied by the correction factor (6) that represents the different DQ alleles.

Table 1. Association of DRB1*13 alleles with acute motor axonal neuropathy (AMAN) or acute inflammatory demyelinating polyneuropathy (AIDP).

	Controls ($n = 34$)	AMAN ($n = 31$)	AIDP ($n = 12$)	P
DRB1*1301	0	6.5	18	.055
DRB1*1302	0	6.5	0	
DRB1*1303	0	3	0	
DRB1*1312	0	3	0	
Total	0	19	18	.009

NOTE. Data are % of subjects or controls with allele.

Discussion

This study suggests that the two forms of GBS that occur in China, AMAN and AIDP, are both characterized by independent HLA associations. Furthermore, each of these forms of GBS has a different HLA association than the other, suggesting a different mechanism of disease development. However, due to the rather small number of cases included in each of the individual groups (12 AIDP, 31 AMAN, and 34 controls), the observed associations should be regarded cautiously. Nevertheless, what is important at this point is that there are certain interesting trends that need to be evaluated carefully in a larger population. Confirmation of any associations between HLA and one of these diseases but not the other will further suggest different pathophysiologic mechanisms underlying AIDP and AMAN. HLA association would additionally suggest an antigen-specific immune response and, therefore, a role for specific T cells.

In a study from England [21], an interesting HLA association (DQB1∗03) was identified among GBS patients with preceding *C. jejuni* infection; however, no HLA association was found among GBS and control subjects. In contrast, in our study, in which GBS patients are classified as AMAN and AIDP subjects, HLA associations are identified between disease status and HLA alleles. It will be very interesting to examine in our population whether the identified HLA associations persist or whether there are new HLA associations identified when each of the disease groups is further divided on the basis of seroconversion status for *C. jejuni* infection. HLA associations will then reflect a complex interaction between disease status infec-

tivity to *C. jejuni* and host factors. We are currently characterizing our subjects for seropositivity to *C. jejuni*.

The precise mechanism by which an antigen-specific T cell–mediated immune response can be explained when the target antigen is believed to be a glycolipid unit is unknown. This will require careful consideration and may eventually open new directions in the way we understand certain aspects of the immune response. Recently, it was shown that monoclonal anti-GM_1 IGM antibodies from peripheral blood lymphocytes of patients with anti-GM_1 antibody-associated neuropathy show diverse binding patterns [26]. It is not unlikely that the reactivity of such monoclonal antibodies extends to nonglycolipids and that the monoclonal antibodies recognize other protein-based structures. Whether the reactivity of an IgM suggests anything about the reactivity of a T cell receptor remains to be seen. Issues of cross-reactivity and mimicry may be important.

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