Commentary

Multiple sclerosis: major histocompatibility complexity and antigen presentation

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Abstract

Multiple sclerosis (MS), like many putative autoimmune diseases, has been known to be associated with the human leukocyte antigen (HLA) class II region for more than 3 decades. However, exactly how HLA class II alleles increase the risk of MS is not yet conclusively known. Recent work in large human cohorts has highlighted the fact that nearly all common *HLA-DRB1* allelotypes are either positively or negatively associated with the disease, detracting from allele-specific antigen presentation as the sole mechanism of MHC associated disease susceptibility. Here, we put into context recent data on the HLA class II region in MS, including allelic heterogeneity, gene-environment interactions and epigenetics. It is clear that a complete understanding of the epistatic interactions and epigenetic features of this region will be crucial to comprehending disease pathogenesis.

Introduction

Multiple sclerosis (MS) as a disease has been recognized for well over a century, but relatively little is understood about its cause. MS is a putative autoimmune disorder of the central nervous system, characterized by inflammatory demyelination, varying degrees of axonal pathology and progressive neurological dysfunction. Risk factors associated with the disease appear to exert effects many years before the clinical onset of MS, lending credence to the idea of a causal cascade in MS development. Genetic-epidemiological studies point unequivocally to large genetic and environmental influences on susceptibility [1]. An association between MS and alleles of the major histocompatibility complex (MHC) was found in the 1970s, notably involving the class II human leukocyte antigen HLA-DR2 [2]. This was later fine-mapped to the extended haplotype *HLA-DRB5*0101-HLA-DRB1*1501-HLA-DQA1*0102-HLA-DQB1*0602* [3] (to briefly explain HLA nomenclature, the first two digits of an allele describe its serological antigen (called an allelotype) while the third and fourth digits are used to list the allele subtypes. Alleles with different numbers in these first four digits must differ by at least one non-synonymous nucleotide substitution).

This extended haplotype confers a relative risk of approximately 3, but much larger effects are seen if haplotypic and diplotypic (both haplotypes in combination) information is taken into account, and the odds ratio for risk spanned by variation in the class II HLA region is thought to exceed 30.

Genome-wide association studies have highlighted the fact that the HLA class II region exerts by far the strongest genetic effect on risk [4], but exactly how it alters the risk of developing MS is not yet fully understood. As HLA-DRB1 alleles have different structural capacities for antigen presentation depending on their amino acid sequence, the MS MHC association has been used to support the concept that disease pathogenesis is the result of an autoimmune reaction, perhaps against myelin-related antigens in the restricting context of *HLA-DRB1*1501*. However, it has become clear only very recently that it is now untenable that all MHC related disease risk is due to the *DRB1*1501* allele, as was originally thought. This conclusion may be unwelcome for those who have made large investments in the transgenic animal models that depend on it, as these models are now clearly uninformative to truly understand disease pathogenesis.

Allelic heterogeneity

While MS is associated with the *HLA-DRB1*1501* haplotype in Northern European populations [3], in other regions like the Mediterranean basin, such as Sardinia, association is predominantly seen with the *HLA-DRB1*0301*, *HLA-DRB1*0405* and *HLA-DRB1*1303* haplotypes [5]. *HLA-DRB1*13* is also MS-associated in Israel [6], but in continental Italy *HLA-DRB1*07* is the primary association [7]. A re-examination of the HLA associations in Northern European MS populations [8-11], using thousands of patients, uncovered many haplotypes (*DRB1*03*, **01*, **10*, **11*, **14*, **08*) that were both positively and negatively associated with the disease. Haplotypes differed in their contribution to disease risk and either acted on their own

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EBV, Epstein-Barr virus; HLA, human leukocyte antigen; MHC, major histocompatibility complex; MS, multiple sclerosis; VDRE, vitamin D response element.

Table 1

Examples of HLA associations with MS across the world among common alleles

HLA-DRB1 allele	Associated population	Approximate odds ratio
*01	Canada, Sweden, UK, US, [8,11,26,27]	0.6
*03 (17)	Canada, Sweden, UK, US, Italy, Sicily, Spain, Sardinia [8-11]	1.7
*04	Sardinia [35]	2.2
*07	Italy [7]	0.6
*08	Canada, UK, US, Italy, Sicily, Spain (15/08 genotype) [8,10,11]	6 (15/08 genotype)
*09	Japan [36]	0.4
*10	Italy, Canada [7,8,11]	2 (protective in Canadians)
*11	Canada, Malta [8,11,37]	0.7
*12†	Canada [11]	0.9
*13	Sardinia, Israel [5,6]	2
*14	Canada, UK, US, Italy, Sicily, Spain [8,10,11]	0.3
*15	Near-universal	3

†Based on a small number of observations. The allele frequency of HLA-DRB1*16 is too low to make any definitive conclusions.

or had an effect *in trans* with another haplotype. Thus, every major allelotype of *HLA-DRB1* is associated with MS (summarized in Table 1).

This conspicuous fact has drawn little attention. Animal models simply transgenic for *HLA-DRB1*1501* seem increasingly irrelevant for the study of the human disease because of it [12]. Indeed, it has recently been shown that *HLA-DRB1*1501* haplotypes can range from super-susceptible to protective depending on other haplotypic features [13]. The *HLA-DRB1* association with MS seems to be geography-dependent and is probably one determinant of the latitude gradient in MS incidence that is seen in temperate climes. It is worth considering that both disease and allele gradients could result from similar environmental pressures. Although associations do reflect the frequency of specific alleles in different countries, the differences among countries cannot completely explain disease frequency. The influence of so many haplotypes on risk, not to mention the prominent interactions, brings into question the venerable belief that MHC associations are determined by structural capacity for antigen presentation.

Structure-function relationships

Different *HLA-DRB1* alleles encode proteins with different binding affinities for disease-related peptides, as determined by their protein sequence. This has plausibly been considered to influence the composition of T cell repertoires, ultimately resulting in *HLA-DRB1* alleles restricting disease risk. However, our analysis [14] has shown that no sequence variant of *HLA-DRB1* can fully explain the risk attributable to all disease-associated alleles across the globe. One explanation could be that disease-causing peptides vary by geography, but the similar disease pathology worldwide would not support this. In the Canadian melting pot of immigrants, MHC associations have remained true to region of origin and give no support to the notion that any geographic specificity of antigenic peptides is relevant (SVR and GCE, unpublished observations).

Environment

Another plausible hypothesis is that the environment of each geographical region interacts with liable *HLA-DRB1* haplotypes. In a given population such interaction could influence the likelihood of presenting disease peptides with a timing and tissue localization that will have an impact on MS susceptibility. This makes the assumption that the associations of MHC class II molecules in MS result entirely from roles in specific and restricted antigen presentation to T cells, a dogma that now warrants reconsideration [15].

Environmental factors with convincing evidence for some involvement in MS pathogenesis include sunshine/vitamin D, Epstein-Barr virus (EBV) and smoking [16-18]. Twin concordance varies by place of birth, strongly hinting that gene-environment interactions will be important in MS [19].

There are several ways in which the environment could interact with the MHC. Recent studies have localized a functional vitamin D response element (VDRE) to the promoter region of *HLA-DRB1* and this VDRE is always present on *HLA-DRB1*15* haplotypes [20]. Although this interaction may have a key role in the increased risk of MS indicated by this haplotype in Northern Europe, it cannot explain why different *HLA-DRB1*15* haplotypes confer different risks [13]. More recently, a second interaction has

been identified involving the curious month-of-birth effect in MS. This has been linked to the same *HLA-DRB1* allele [21].

No studies have yet examined the role of smoking-HLA interactions in MS. Investigations of anti-EBV antibody levels or symptomatic infection with EBV, *HLA-DRB1*15* and the risk of MS have shown that *HLA-DRB1*15* may act synergistically with anti-EBV antibodies or infectious mononucleosis to increase MS risk [22,23]. The biological nature of this statistical interaction needs to be elucidated, but again it must be remembered that *HLA-DRB1*15* is not the only MS risk allele.

Epistasis or haplotype effects

Although other risk components are present on *HLA-DRB1* haplotypes in the class II region, and *HLA-DQ* molecules undoubtedly have a role [5,24], there is no single *HLA-DQ* element common to all disease-associated haplotypes. It does, however, seem that there are combinations of HLA-DQB1, HLA-DQA1 and HLA-DRB1 that are required to confer risk of MS [25], and investigation of alleles present at *HLA-DQ* have shed light on haplotypic associations of *HLA-DRB1*13* and *HLA-DRB1*04* in MS [25]. These haplotypic effects may reflect the effects of selection for functions that are epistatic in nature. HLA class I haplotype tagging can differentiate the risk conferred by different *HLA-DRB1*15* haplotypes (despite all having the same alleles of DQ) [13], further indicating that there is more in the MHC than *HLA-DQ* and *HLA-DRB1* in determining MS risk. HLA class I may be an epistatic partner of *HLA-DRB1,* but given that several class I alleles differentiate *HLA-DRB1*15* haplotypes [13] and that HLA class I associations in MS have been conflicting (HLA-A, B and C have all been implicated [26-28]), it is unlikely that HLA class I has a major role in MS, and the more reliable haplotype transmission data imply it is not an independent contributor to risk [29].

Epigenetics

A missing link seems to be the epigenetic modification of class II region genes. The genetic epidemiology of MS had clearly implied a major epigenetic effect, with mothers more likely to be the common parent in affected half-siblings [18] and to be the intervening parent when affected aunt-niece pairs are studied [30]. This effect has now been localized to the MHC itself [31]. DNA and chromatin modifications regulate the expression of HLA class II genes [32], and the epigenetic status of the genome varies dynamically compared with the static DNA sequence and is influenced by the environment [33]. MS environmental factors (vitamin D, smoking, EBV) can all influence the epigenome [1]. It is therefore plausible that the different HLA associations observed across the globe are a reflection of specific environmental factors influencing epigenetic marks on liable haplotypes, which affect the expression or function of class II genes and permit the MS pathogenic cascade. Epigenetics may be the mechanism that brings together many of the factors (genetic and environmental) that are MS-associated. Epigenetics has been suggested to underlie recombination hotspots [34] and this may provide an additional explanation for the fact that linkage disequilibrium maintains particular haplotypic combinations in the class II region. Combining epigenetic information with class II haplotype sequence will probably provide an improved understanding of MS disease mechanisms.

This brings us back to the venerable concept of antigen presentation as an explanation for MHC class II disease associations. The data so far are inconclusive, but it may be time to recall that many of the concepts of immune response genes came from very restricted experimental situations. It is not a given that the frequently much more complex circumstance of autoimmune disease would be analogous. Many putative autoimmune diseases lack even a single validated autoantigen. The paradigm for MHC-disease association continues to be MHC class II allele-specific antigen presentation to T cells. However, MS suggests a broader view, with other features of the haplotypes, including epigenetic modifications, appearing to participate in important epistatic interactions. The sheer variety of disease-associated alleles in this and other autoimmune diseases warrants reconsideration of the paradigm. It may be that MHC disease associations are driven less by allele-specific antigen presentation and more by the propensity of specific haplotypes to undergo strategic epigenetic modifications. The role of DNA methylation in the process of tissue-specific expression might plausibly relate to the establishment of immunological tolerance, but there is no direct evidence to support such a notion.

Conclusions

The notion of *HLA-DRB1*1501* as the one disease allele in MS is rapidly yielding to a more complex view. An orchestra of class II genes, their interactions and their regulatory components have now been shown to be important. The epigenetic pattern within the MHC laid down by differential methylation warrants consideration as the master conductor of MHC diplotype-associated disease risk.

Competing interests

The authors declare that they have no competing interests.

Authors' contributions

SVR and GCE conceived the idea of the commentary and wrote the manuscript.

Authors' information

GCE is the Action Research Professor of Clinical Neurology at the University of Oxford. He initiated and leads the Canadian Collaborative Project on the Genetic Susceptibility to Multiple Sclerosis (CCPGSMS). Through the CCPGSMS he conducted much of the work that

identified the importance and identity of genetic factors in MS and delineated the natural history of the disease. SVR is a Junior Research Fellow at Somerville College, University of Oxford and a Goodger Scholar at the University of Oxford. His interest lies in how epistasis and gene-environment interactions at the HLA region alter susceptibility to MS.

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