Multiple sclerosis: major histocompatibility complexity and antigen presentation

Sreeram V Ramagopalan**† and George C Ebers**†

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 alleles 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, genetic-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

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

---

### 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.
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.

References

1. Ramagopalan SV, Dyment DA, Ebers GC: Genetic epidemiology: the use of old and new tools for multiple sclerosis. Trends Neurosci 2008, 31:645-652.
2. Jersild C, Fog T, Hansen GS, Thomsen M, Sveigaard A, Dupont B: Histocompatibility determinants in multiple sclerosis, with special reference to clinical course. Lancet 1973, 2:1221-1225.
3. Fogdell A, Hillert J, Sachs C, Olerup O: The multiple sclerosis- and narcolepsy-associated HLA class II haplotype includes the DRB5*0101 allele. Tissue Antigens 1995, 46:333-336.
4. Haffer DA, Compston A, Sawyer S, Lander ES, Daly MJ, De Jager PL, de Bakker PI, Gabriel SB, Mirel DB, Iverson AJ, Pericak-Vance MA, Gregory SG, Rioux JD, McAuley JL, Haines JL, Barcellos LF, Cree B, Okenberg JR, Hauser SL: Risk alleles for multiple sclerosis are identified by a genome-wide study. N Engl J Med 2007, 357:851-862.
5. Marrosu MG, Murr U, Murr U MR, Costa G, Zavattari P, Whalen M, Cocco E, Mancosu C, Schirru L, Solla E, Fadda E, Melis C, Porru I, Roleus M, Cucca F: Dissection of the HLA association with multiple sclerosis in the founder isolated population of Sardinia. Hum Mol Genet 2001, 10:2979-2991.
6. Kwon OJ, Kariu A, Israel S, Brautbar C, Amar A, Meiner Z, Abramsky O, Karussis D: The inheritance of resistance alleles in multiple sclerosis by latitude of birthplace. Ann Neurol 2007, 61:504-515.
7. Ballerini C, Guerini FR, Rombola G, Rosati E, Massacesi L, Urbinati M, Sulli A, Rizzardi G, Cavalleri L, Novelli M, Chiodo M, Alizadeh M, Momigliano-Richiardi P, D’Alfonso S: Differential twin concordance for multiple sclerosis susceptibility: accounting for linkage disequilibrium. Ann Neurol 2007, 62:56-64.
8. Kwon OJ, Karni A, Israel S, Brautbar C, Amar A, Meiner Z, Abramsky O, Karussis D: The inheritance of resistance alleles in multiple sclerosis. Part I: the role of infection. Acta Neurol Scand 2007, 115:306-311.
9. Lincol MR, Ramagopalan SV, Chao MJ, Herrera BM, Deluca GC, Orton SM, Dyment DA, Sadovnick AD, Ebers GC: Complex interactions among MHC haplotypes in multiple sclerosis susceptibility and resistance. Hum Mol Genet 2005, 14:2019-2026.
10. Masterman T, Ligers A, Olsson T, Andersson M, Olerup O, Hillert J: HLA-DR15 is associated with lower age at onset in multiple sclerosis. Ann Neurol 2000, 48:211-219.
11. Barcellos LF, Sawyer S, Ramsay PP, Baranzini SE, Thomson G, Briggs F, Cree BC, Begovich AB, Villoslada P, Montalban X, Uccelli A, Savettieri G, Lincol RR, Deloa C, Haines JL, Pericak-Vance MA, Compston A, Hauser SL, Okenberg JR: HLA-DRB1 locus determines multiple sclerosis susceptibility. Proc Natl Acad Sci USA 2008, 105:7542-7547.
12. Brynedal B, Duvefelt K, Jonasdottir G, Roos IM, Akesson E, Palmgren J, Hillert J: HLA-A confers an HLA-DRB1 independent influence on the risk of multiple sclerosis. PLoS ONE 2007, 2:e664.
13. Yeo TW, De Jager PL, Gregory SG, Barcellos LF, Walton A, Goris A, Fenoglio C, Ban M, Taylor CJ, Goodman RS, Walsh E, Wolfish CS, Horton R, Traherne J, Beck S, Trowsdale J, Cailler SJ, Ivison AJ, Green T, Pabywajlo S, Lander ES, Pericak-Vance MA, Haines JL, Daly MJ, Okenberg JR, Hauser SL, Compston A, Haffer DA, Rioux JD, Sawyer S: A second major histocompatibility complex susceptibility locus for multiple sclerosis. Ann Neurol 2007, 61:228-238.
14. Madsen LS, Andersson EC, Janssen L, Krogsgaard M, Andersen CB, Engberg J, Strominger JL, Sveigaard A, Jhorth JP, Holmdahl R, Wucherpfennig KW, Fugger L: A humancized model for multiple sclerosis using HLA-DR2 and a human T-cell receptor. Nat Genet 1999, 23:343-347.
15. Chao MJ, Barnardo MC, Lincoln MR, Ramagopalan SV, Herrera BM, Dyment DA, Montpetit A, Sadovnick AD, Knight JC, Ebers GC: HLA class II alleles tag HLA-DRB1*1501 haplotypes for differential risk in multiple sclerosis susceptibility. Proc Natl Acad Sci USA 2008, 105:13069-13074.
16. Ramagopalan SV, McMahan R, Dyment DA, Sadovnick AD, Ebers GC, Wittkowski KM: An extension to a statistical approach for family based association studies provides insights into genetic risk factors for multiple sclerosis in the HLA-DRB1 gene. BMC Med Genet 2009, 10:10.
17. Hiremath MM, Chen VS, Suzuki K, Ting JP, Matsushima GK: MHC class II exacerbates demyelination in vivo independently of T cells. J Neuroimmunol 2008, 203:23-32.
18. Ascherio A, Munger KL: Environmental risk factors for multiple sclerosis. Part II: noninfectious factors. Ann Neurol 2007, 61:297-307.
19. Islam T, Gauderman WJ, Cozen W, Hamilton AS, Burnett ME, Mack TM: Differential twin concordance for multiple sclerosis by latitude of birthplace. Ann Neurol 2006, 60:56-64.
20. Ramagopalan SV, Maugeri NJ, Handunnetthi L, Lincoln MR, Orton SM, Dyment DA, Deluca GC, Herrera BM, Chao MJ, Sadovnick AD, Ebers GC, Knight JC: Expression of the multiple sclerosis-associated MHC class II Allele HLA-DRB1*1501 is regulated by vitamin D. PLoS Genet 2009, 5: e1000369.
21. Ramagopalan SV, Link J, Byrnes JK, Dyment DA, Giovannoni G, Hintzen RQ, Sundqvist E, Kockum I, Smestad C, Lie BA, Harbo HF, Padyukov L, Alfredsson L, Olsson T, Sadovnick AD, Hillert J, Ebers G: HLA-DRB1 and month-of-birth in multiple sclerosis. Neurology in press.
22. De Jager PL, Simon KC, Munger KL, Rioux JD, Haffer DA, Ascherio A: Integrating risk factors: HLA-DRB1*1501 and Epstein-Barr virus in multiple sclerosis. Neurology 2008, 70:1113-1118.
23. Nielsen TR, Rostgaard K, Asling J, Steffensen R, Oturai A, Jersild C, Koch-Henriksen N, Sorensen PS, Hjalgrim H: Effects of infectious mononucleosis and HLA-DRB1*15 in multiple sclerosis. Mult Scler 2009, 15:431-436.
24. Alves-Leon SV, Papais-Alvarenga R, Magalhaes M, Alvarenga M, Thuler LC, Fernandez y Fernandez O: Ethnicity-dependent association of HLA DRB1-DQA1-DQB1 alleles in Brazilian multiple sclerosis patients. Acta Neurol Scand 2007, 115:306-311.
25. Lincoln MR, Ramagopalan SV, Chao MJ, Herrera BM, Deluca GC, Orton SM, Dyment DA, Sadovnick AD, Ebers GC: Epistasis among HLA-DRB1, HLA-DQA1, and HLA-DQB1 loci determines multiple sclerosis susceptibility. Proc Natl Acad Sci USA 2008, 105:7542-7547.
26. Brynedal B, Duvefelt K, Jonasdottir G, Roos IM, Akesson E, Palmgren J, Hillert J: HLA-A confers an HLA-DRB1 independent influence on the risk of multiple sclerosis. PLoS ONE 2007, 2:e664.
27. Yeo TW, De Jager PL, Gregory SG, Barcellos LF, Walton A, Goris A, Fenoglio C, Ban M, Taylor CJ, Goodman RS, Walsh E, Wolfish CS, Horton R, Traherne J, Beck S, Trowsdale J, Cailler SJ, Ivison AJ, Green T, Pabywajlo S, Lander ES, Pericak-Vance MA, Haines JL, Daly MJ, Okenberg JR, Hauser SL, Compston A, Haffer DA, Rioux JD, Sawyer S: A second major histocompatibility complex susceptibility locus for multiple sclerosis. Ann Neurol 2007, 61:228-238.
28. Lorentzen AR, Karlsen TH, Olsson M, Smestad C, Mero IL, Woldseth B, Sun JY, Senitzer D, Celius EG, Thorsby E, Spurkland A, Lie BA, Harbo HF: Killer immunoglobulin-like receptor ligand HLA-Bw4 protects against multiple sclerosis. Ann Neurol 2009, 65:688-688.
29. Chao MJ, Barnardo MC, Lui GZ, Lincoln MR, Ramagopalan SV, Herrera BM, Dyment DA, Sadovnick AD, Ebers GC: Transmission of class I/II multi-locus MHC haplotypes and multiple sclerosis susceptibility: accounting for linkage disequilibrium. Hum Mol Genet 2007, 16:1951-1958.
30. Lincoln MR, Ramagopalan SV, Chao MJ, Sadovnick AD, Ebers GC: Parent-of-origin effects in MS. Observations from avuncular pairs. Neurology 2008, 71:799-803.
31. Chao MJ, Ramagopalan SV, Herrera BM, Lincoln MR, Dyment DA, Sadovnick AD, Ebers GC: Epigenetics in multiple
sclerosis susceptibility: difference in transgenerational risk localizes to the major histocompatibility complex. *Hum Mol Genet* 2009, 18:261-266.

32. Gialitakis M, Kretsovali A, Spilianakis C, Kravariti L, Mages J, Hoffmann R, Hatzopoulos AK, Papamatheakis J: Coordinated changes of histone modifications and HDAC mobilization regulate the induction of MHC class II genes by Trichostatin A. *Nucleic Acids Res* 2006, 34:765-772.

33. Feinberg AP: Epigenetics at the epicenter of modern medicine. *JAMA* 2008, 299:1345-1350.

34. Neumann R, Jeffreys AJ: Polymorphism in the activity of human crossover hotspots independent of local DNA sequence variation. *Hum Mol Genet* 2006, 15:1401-1411.

35. Marrosu MG, Murru MR, Costa G, Cucca F, Solgiu S, Rosati G, Muntoni F: Multiple sclerosis in Sardinia is associated and in linkage disequilibrium with HLA-DR3 and -DR4 alleles. *Am J Hum Genet* 1997, 61:454-457.

36. Matsuoka T, Matsushita T, Osoegawa M, Kawano Y, Minohara M, Mihara F, Nishimura Y, Ohyagi Y, Kira J: Association of the HLA-DRB1 alleles with characteristic MRI features of Asian multiple sclerosis. *Mult Scler* 2008, 14:1181-1190.

37. Dean G, Yeo TW, Goris A, Taylor CJ, Goodman RS, Elian M, Galea-Debono A, Aquilina A, Felice A, Vella M, Sawcer S, Compton DA: HLA-DRB1 and multiple sclerosis in Malta. *Neurology* 2007, 70:101-105.

Published: 6 November 2009
doi:10.1186/gm105
© 2009 BioMed Central Ltd