Anne C. La Flamme
Jacqueline Monique Orian *Editors*

Emerging and Evolving Topics in Multiple Sclerosis Pathogenesis and Treatments

Springer

Current Topics in Behavioral Neurosciences

Volume 26

About this Series

Current Topics in Behavioral Neurosciences provides critical and comprehensive discussions of the most significant areas of behavioral neuroscience research, written by leading international authorities. Each volume offers an informative and contemporary account of its subject, making it an unrivalled reference source. Titles in this series are available in both print and electronic formats.

With the development of new methodologies for brain imaging, genetic and genomic analyses, molecular engineering of mutant animals, novel routes for drug delivery, and sophisticated cross-species behavioral assessments, it is now possible to study behavior relevant to psychiatric and neurological diseases and disorders on the physiological level. The *Behavioral Neurosciences* series focuses on "translational medicine" and cutting-edge technologies. Preclinical and clinical trials for the development of new diagnostics and therapeutics as well as prevention efforts are covered whenever possible.

More information about this series at http://www.springer.com/series/7854

Anne C. La Flamme
Jacqueline Monique Orian
Editors

Emerging and Evolving Topics in Multiple Sclerosis Pathogenesis and Treatments

Springer

Editors
Anne C. La Flamme
School of Biological Sciences, Malaghan
   Institute of Medical Research
Victoria University of Wellington
Wellington
New Zealand

Jacqueline Monique Orian
Department of Biochemistry, School of
   Molecular Sciences
La Trobe Institute for Molecular Science
Melbourne
Australia

Preface

Over the past decade, we have made great advances in the field of multiple sclerosis (MS) research. While some of these advances have been through new approaches and ideas that have emerged in the last decade such as the newly identified protective role that amyloid proteins may play in MS (Kurnellas et al. 2015), others have evolved from previous theories and ideas that have only now gained momentum and a deeper understanding. This book has been written to cover these emerging and evolving topics and to highlight the substantial advancements made in understanding the factors regulating susceptibility or disease progression, identifying new ways to monitor or predict MS pathology, or developing new strategies for treating MS.

This book begins with three chapters that explore recent advances in our understanding of how genetic factors regulate MS susceptibility and disease progression including HLA (Greer 2014) and sex-related factors (Dunn et al. 2015a, b). Greer discusses in depth the known associations with particular HLA alleles, both detrimental and protective, and highlights the significant association of HLA-DRB1*1501 with MS in Caucasians (Greer 2014). For other ethnic populations, different alleles appear to contribute to MS susceptibility and these different ethnic associations are presented (Greer 2014). While the association between HLA and MS susceptibility has been well documented after its first reports in 1976 (Compston et al. 1976; Terasaki et al. 1976), it is only in recent years that the effect of HLA on disease subtype and progression has been firmly delineated, in part due to advances in DNA sequencing and genetic analyses. However, despite the identification of other genetic loci that associate with MS susceptibility (Beecham et al. 2013; Hafler et al. 2007; Bahlo et al. 2009), HLA remains the major genetic factor, and the mechanism by which it regulates susceptibility is an exciting and evolving area of research. Consequently, Greer describes a novel mechanism by which these different alleles may facilitate the presentation of myelin peptides to induce the autoimmune T cell responses that drive immune-mediated demyelination in MS (Greer 2014).

In addition to the strong association between HLA and MS is the clear gender bias in MS where females are 3 times more likely to develop MS than males (Sellner et al. 2011). While this gender bias has been recognized for decades, significant research has been undertaken to understand what drives this bias and how gender affects MS onset and progression (Sellner et al. 2011; Orton et al. 2006). Dunn et al. review recent advances in these areas in two separate chapters (Dunn et al. 2015a, b). The first chapter details in depth the biology of how immune responses (T and B cell), microglial responses, and blood brain barrier permeability differ between men and women (Dunn et al. 2015a). Additionally, the protective or detrimental involvement of sex hormones is explored as an explanation for the more robust autoimmune responses measured in females (Dunn et al. 2015a). This review combines the results from epidemiological studies, clinical trials, and research using animal models of MS such as experimental autoimmune encephalomyelitis (EAE) and concludes that these studies support the increased susceptibility of females to MS in part due to enhanced autoimmune response induction in the early phase of disease compared to males (Dunn et al. 2015a).

The second chapter by Dunn et al. takes a different approach and investigates the relationship between gender and disease incidence and progression (Dunn et al. 2015b). To understand the impact of gender on disease incidence, Dunn et al. explore the interaction of gender with known environmental or lifestyle factors such as Epstein-Barr virus (EBV) infection, smoking, sunlight, and vitamin D (Dunn et al. 2015b). While these environmental factors are discussed in greater depth by Hedström et al. (2015), this chapter describes preliminary evidence that infectious mononucleosis (caused by EBV), sunlight and vitamin D but not smoking have a stronger association between MS and women than men, but Dunn et al. caution that further research in this area is required to understand fully the relationship between gender, environmental factors, and MS onset (Dunn et al. 2015b). Additionally, this chapter reviews recent discoveries in how gender affects disease progression and clearly segregates the distinct effects observed on disability, cognitive decline, white and grey matter pathology, and remyelination. Interestingly, these studies suggest that men, not women, appear to show a more rapid disease progression as measured by disability and cognitive function (Dunn et al. 2015b). Finally, Dunn et al. conclude by pulling together how sex hormones may be contributing to these changes in MS progression by directly altering neuroprotection as opposed to autoimmune response induction and development as discussed in Dunn et al. (2015a). Indeed, these studies suggest a positive effect of testosterone and estriol on cognitive performance but differing effects on lesion reduction (decreased by estriol) and atrophy (decreased by testosterone) (Dunn et al. 2015b). While exciting, the results from clinical trials need to be cautiously interpreted due to the small number of participants, but certainly support the need for further study to elucidate the biological processes mediating how sex hormones are involved in MS pathology.

Although heritable factors such as HLA and sex are clearly strong regulators of MS susceptibility, previous research has also highlighted the important involvement of the environment and lifestyle in determining if MS develops in susceptible

individuals. Hedström et al. provide an excellent overview of the recent advances in this area and focus on individual environmental or lifestyle factors (e.g. sunlight, vitamin D, smoking, EBV infection) and how they interact with the major genetic factor (i.e. HLA) (Hedström et al. 2015). Sunlight was proposed as a protective factor in 1960 by Sir Donald Acheson (Acheson et al. 1960) and the mechanism of protection was proposed to be due to vitamin D. Although sunlight and vitamin D have not been shown to interact with HLA genes (Hedström et al. 2015), they are clearly related to each other, and Pakpoor et al. provide an in-depth review of recent studies investigating how vitamin D is protective (Pakpoor and Ramagopalan 2014).

However, our previous understanding that sunlight was protective solely through its role in vitamin D production needs to be revised, and in the chapter by Marsh-Wakefield and Byrne, they clearly outline the vitamin D-independent protective effects of sunlight (Marsh-Wakefield and Byrne 2015). Furthermore, they delve into the immunological mechanisms by which ultraviolet B light (UVB) may regulate and suppress autoimmune response development (Marsh-Wakefield and Byrne 2015). To elucidate the protective immune pathways induced by UVB, March-Wakefield and Byrne detail recent studies that use the EAE rodent model of MS to define the specific involvement of innate and adaptive immune cells in EAE after UVB exposure. In particular, these studies indicate that UVB induces the release of soluble factors such as platelet-activating factor, serotonin, interleukin (IL)-33, IL-10, which in turn lead to the activation and recruitment of regulatory immune cell subsets that can suppress autoimmune response induction (Marsh-Wakefield and Byrne 2015). Marsh-Wakefield and Byrne describe the evidence that regulatory T cells and/or regulatory B cells are induced by UVB exposure, yet the relevance of these regulatory cells and pathways to MS protection by UVB remains to be confirmed (Marsh-Wakefield and Byrne 2015).

Hedström et al. also discuss other environmental and lifestyle factors that show a strong interaction with HLA genes including EBV infection, smoking, and obesity, and while these factors are associated with an elevated MS risk independent of HLA, the risk is significantly elevated by HLA (Hedström et al. 2015). For example, although HLA genes only modestly increase the odds ratio due to EBV serology from 13.5 to 16, they dramatically increase the odds ratio due to active smoking from 1.6 to 15 (Hedström et al. 2015). Together these chapters offer critical insight into our current understanding of the lifestyle and environmental factors involved in regulating MS susceptibility (Hedström et al. 2015; Pakpoor and Ramagopalan 2014; Marsh-Wakefield and Byrne 2015). It is hoped that armed with this knowledge, more informed lifestyle choices (e.g. smoking, obesity) can be made by persons, who have a genetic susceptibility to MS. Additionally, these studies highlight potential intervention strategies such as EBV vaccination or prophylactic vitamin D administration that could lower the risk of MS irrespective of genetic susceptibility and HLA, in particular.

Key to the advancement of MS research has been the development of tools to enable a deeper interrogation of the specific pathology that arises in MS. Over the years several different animals models have been developed including the EAE

model, Theiler's encephalomyelitis virus, and the cuprizone model of non-immune demyelination (Steinman 1999). Although no single model recapitulates all aspects of MS disease progression and pathology, each is able to model specific aspects of the disease. For detailed and mechanistic investigation of the immunological processes driving MS, the EAE mouse model is the most widely used, but this model is highly dependent upon the inbred strain and myelin antigen used. The chapter by Dang et al. describes this model and details the disease characteristics in several commonly used mouse strains (Dang et al. 2015). Additionally, Dang et al. provide evidence that the NOD/Lt strain immunized with myelin oligodendrocyte glycoprotein induces a chronic relapsing disease with upper central nervous system (CNS) involvement unlike many of the current EAE mouse models that either display a monophasic disease or develop only lower CNS lesions (Dang et al. 2015). Given that lesions occur in both the grey and white matter with early axonal injury and appear to recapitulate certain MS lesion subtypes, the EAE variant provides a new tool to address fundamental questions about MS pathogenesis and investigate the potential of new MS treatments (Dang et al. 2015).

In concert with the development of new tools to model disease as described by Dang et al. (2015) is the development of technologies that enable a better understanding of the disease process in humans. To this end, Gnanapavan and Giovannoni review the current biomarkers that have been associated with MS neurodegeneration and place a specific emphasis on the development of neurofilaments as predictors of disability (Gnanapavan and Giovannoni 2014). What clearly arises from this review is an appreciation that biomarker discovery and validation is a difficult path but one that can reap enormous rewards in terms of disease insight and valuable surrogate outcomes for trials for new MS therapies (Gnanapavan and Giovannoni 2014).

Over the past decade a wide variety of new therapeutics have been developed to treat MS, and many more are in the pipeline. While some of these therapies have been developed specifically for MS, others have had been developed and used in other diseases before being applied successfully to MS. One such therapy, which is still under investigation, is helminth therapy, which was successfully developed for inflammatory bowel diseases (Fleming and Weinstock 2015). Based on the "Old Friends Hypothesis," extensive preclinical research has demonstrated that helminth infection can be protective against a variety of inflammatory diseases including MS (Fleming and Weinstock 2015). Tanasescu and Constantinescu review the current status of this research and the on-going trials using live helminths in relapsing-remitting MS patients (Tanasescu and Constantinescu 2014). While it is evident that, like many other therapies that work in animal models, the efficacy of helminth therapy in humans is not as clear-cut, the preliminary trials have shown a good safety profile and encouraging results suggesting that our old friends may provide a new treatment (Tanasescu and Constantinescu 2014).

The book concludes by reviewing recent research demonstrating a protective role for amyloid fibril-forming peptides and proteins in the EAE mouse model that on the face appears to directly contradict to the proposed detrimental role for amyloid proteins in neurodegenerative diseases such as Alzheimer's disease or

Parkinson's disease (Kurnellas et al. 2015). Kurnellas et al. discuss current research investigating how these proteins and peptides exert their protective effects and present compelling evidence suggesting that they act in an anti-inflammatory manner (Kurnellas et al. 2015). Given that administration of the amyloid fibril-forming peptides was therapeutic, Kurnellas et al. suggest that this pathway may provide novel therapeutic approach for the treatment of neuroinflammatory conditions such as MS (Kurnellas et al. 2015).

Together these chapters showcase many of the emerging and evolving topics in MS and provide in depth discussions of how these advances in our understanding of MS pathogenesis will lead to better therapies, diagnosis or even prevention. Ultimately, to be useful these advances must now be taken up by the neurologists who treat MS patients such that they can modify and improve their clinical practice.

Wellington, New Zealand   Anne C. La Flamme
Melbourne, Australia   Jacqueline Monique Orian

References

Acheson ED, Bachrach CA, Wright EM (1960) Some comments on the relationship of the distribution of multiple sclerosis to latitude, solar radiation, and other variables. Acta Psychiatr Scand 35(Suppl 147):132–147

Beecham AH, Patsopoulos NA, Xifara DK, Davis MF, Kemppinen A et al (2013) Analysis of immune-related loci identifies 48 new susceptibility variants for multiple sclerosis. Nat Genet 45:1353–1360

Compston DA, Batchelor JR, McDonald WI (1976) B-lymphocyte alloantigens associated with multiple sclerosis. Lancet 2:1261–1265

Dang PT, Bui Q, D'Souza CS, Orian JM (2015a) Modelling MS: Chronic-Relapsing EAE in the NOD/Lt Mouse Strain. Curr Top Behav Neurosci

Dunn SE, Gunde E, Lee H (2015b) Sex-Based Differences in Multiple Sclerosis (MS): Part II: Rising Incidence of Multiple Sclerosis in Women and the Vulnerability of Men to Progression of this Disease. Curr Top Behav Neurosci

Dunn SE, Lee H, Pavri FR, Zhang MA (2015) Sex-Based Differences in Multiple Sclerosis (Part I): Biology of Disease Incidence. Curr Top Behav Neurosci

Fleming JO, Weinstock JV (2015) Clinical trials of helminth therapy in autoimmune diseases: rationale and findings. Parasite Immunol 37:277–292

Bahlo et al (2009) Genome-wide association study identifies new multiple sclerosis susceptibility loci on chromosomes 12 and 20. Nat Genet 41:824–828

Gnanapavan S, Giovannoni G (2014) Developing Biomarkers for MS. Curr Top Behav Neurosci

Greer JM (2014) The Role of HLA in MS Susceptibility and Phenotype. Curr Top Behav Neurosci

Hafler DA, Compston A, Sawcer S, Lander ES, Daly MJ, De Jager PL, de Bakker PI, Gabriel SB, Mirel DB, Ivinson AJ, Pericak-Vance MA, Gregory SG, Rioux JD, McCauley JL, Haines JL, Barcellos LF, Cree B, Oksenberg JR, Hauser SL (2007) Risk alleles for multiple sclerosis identified by a genomewide study. N Engl J Med 357:851–862

Hedström AK, Olsson T, Alfredsson L (2015) The Role of Environment and Lifestyle in Determining the Risk of Multiple Sclerosis. Curr Top Behav Neurosci

Kurnellas MP, Rothbard JB, Steinman L (2015) Self-Assembling Peptides Form Immune Suppressive Amyloid Fibrils Effective in Autoimmune Encephalomyelitis. Curr Top Behav Neurosci

Marsh-Wakefield F, Byrne SN (2015) Photoimmunology and Multiple Sclerosis. Curr Top Behav Neurosci

Orton SM, Herrera BM, Yee IM, Valdar W, Ramagopalan SV, Sadovnick AD, Ebers GC (2006) Sex ratio of multiple sclerosis in Canada: a longitudinal study. Lancet Neurol 5:932–936

Pakpoor J, Ramagopalan S (2014) Evidence for an Association Between Vitamin D and Multiple Sclerosis. Curr Top Behav Neurosci

Sellner J, Kraus J, Awad A, Milo R, Hemmer B, Stuve O (2011) The increasing incidence and prevalence of female multiple sclerosis–a critical analysis of potential environmental factors. Autoimmun Rev 10:495–502

Steinman L (1999) Assessment of animal models for MS and demyelinating disease in the design of rational therapy. Neuron 24:511–514

Tanasescu R, Constantinescu CS (2014) Helminth Therapy for MS. Curr Top Behav Neurosci

Terasaki PI, Park MS, Opelz G, Ting A (1976) Multiple sclerosis and high incidence of a B lymphocyte antigen. Science 193:1245–1247

Contents

The Role of HLA in MS Susceptibility and Phenotype

Judith M. Greer

Abstract One of the most consistent findings in multiple sclerosis (MS) is that development of MS is linked with carriage of the class II human leucocyte antigen (HLA) molecule HLA-DRB1*15:01; around 60 % of Caucasian MS patients carry this allele compared to 25–30 % of ethnically matched healthy individuals. However, other HLA molecules have also been linked to the development of MS. In this chapter, the association between different HLA types and susceptibility to MS will be reviewed, and other linkages between the carriage of specific HLA molecules and clinical and experimental findings in MS will be considered.

Keywords Human leucocyte antigen (HLA) · Multiple sclerosis (MS) · Disease susceptibility · Clinical phenotypes

Contents

J.M. Greer (✉)
The University of Queensland, UQ Centre for Clinical Research,
Building 71/918 Riyal Brisbane and Women's Hospital, Brisbane, QLD 4029, Australia
e-mail: j.greer@uq.edu.au

1 HLA Molecules

Human leucocyte antigen (HLA) is the designation used for the major histocompatibility (MHC) molecules found in humans, and the HLA gene region can be divided into class I, class II and class III. This review will focus only on class I and class II HLA molecules that are expressed at the cell surface. Many subtle differences in the sequences of the HLA molecules exist, and the nomenclature of HLA molecules has evolved over time to accommodate this complexity; an additional layer of complexity in interpreting HLA data comes from the flexible use of either generic names (e.g. HLA-A2 or HLA-DR4) or DNA genotyping nomenclature. A list of recognized genotypes and recognized generic and serological names is published every few years (Holdsworth et al. 2008). The generic names are determined using either serological phenotyping of HLA or low-resolution DNA genotyping, but they do not distinguish closely related alleles, e.g. HLA-A2 could include any one of over 100 HLA-A*02 alleles or subtypes. The genotyping nomenclature shows the particular HLA gene locus, the allele group (which is usually related to the generic group) and the specific HLA allele (e.g. HLA-A*02:01 or HLA-DRB1*04:07).

Class I HLA molecules are constitutively expressed on most cells in the body, and present peptides are derived from the cytosol (e.g. viral or tumour peptides) to CD8$^+$ T cells. The class I HLA molecules are heterodimers formed from an α chain and the β2-microglobulin (β2 m) subunit (Fig. 1). The α chains of the class I HLA molecules are encoded within the MHC gene cluster on chromosome 6 and are highly polymorphic (particularly the most highly expressed class I types, HLA-A, HLA-B and HLA-C—see Table 1). In contrast, β2 m is encoded outside of the MHC on chromosome 15, and humans have only 3 alleleic variants of β2 m.

The class II HLA molecules are expressed primarily on antigen-presenting cells (APCs: dendritic cells, macrophages and B cells), although some other cell types can upregulate class II HLA under inflammatory conditions. Class II HLA molecules present peptides that are derived from proteins progressing through the endosomal/lysosomal pathway (i.e. proteins that have typically been internalized by the APCs) to CD4$^+$ T cells. Class II HLA molecules are formed from an α chain and a β chain (Fig. 1), both of which are encoded within the MHC gene cluster. The α chain of HLA-DR is not very polymorphic, but the α chains of HLA-DQ and HLA-DP are polymorphic, and the β chains of all subtypes are highly polymorphic (Table 2). The HLA-DR gene cluster contains an additional β chain gene, the product of which can also pair with a DRα chain to form DRB3, DRB4 or DRB5 molecules (which are also polymorphic—see Table 2). Expression of these DRB3, DRB4 or DRB5 molecules is linked to specific clusters of DRB1 molecules, designated as DR1, DR51, DR52, DR53 and DR8 clusters (Fig. 2).

One allele of each HLA subtype is inherited from each parent; therefore, for example each person has at least 3 (if they inherit identical alleles from each parent) and up to 8 (if they inherit different DR51, DR52 or DR53 clusters from each parent) class II HLA molecules expressed on the surface of their antigen-presenting cells. Due to strong linkage disequilibrium within the MHC region, specific ancestral