Available online at www.sciencedirect.com
ScienceDirect
Autoimmunity Reviews 5 (2006) 532–536
AUTOIMMUNITY REVIEWS
www.elsevier.com/locate/autrev

A model for the comprehensive investigation of a chronic autoimmune disease: The multiple sclerosis CLIMB study

Susan A. Gauthier [a], Bonnie I. Glanz [a], Micha Mandel [b], Howard L. Weiner [a,*]

[a] *Partners Multiple Sclerosis Center, Brigham and Women's Hospital, Harvard Medical School, 333 Longwood Avenue, Boston, MA 02115, United States*
[b] *Harvard School of Public Health, Department of Biostatistics, Boston, MA, United States*

Available online 22 March 2006

Abstract

Multiple sclerosis (MS) is a chronic inflammatory disease of central nervous system (CNS) resulting in various disabilities including weakness and imbalance, visual abnormalities, changes in cognition, as well as bladder and sexual dysfunction. The majority of patients begin with a relapsing–remitting course of the disease until eventually there is a progressive decline in disability. With FDA-approved disease modifying therapy now given to the majority of MS patients early in the course of the disease, the advent of MRI imaging, as well as advances in immunology and genetics, the study of MS has entered into an exciting era. Natural history studies of untreated patients have provided a guide for disease prognosis based on the clinical features of the disease but have limited utility in this new era of MS. Major questions are unanswered, including how does treatment affect the long-term clinical course of the disease and are there major subcategories of the disease with different implications for treatment and outcome. Advances in our ability to clinically measure and monitor the disease through MRI imaging technology, immunology, and genetic analysis provide the opportunity to address these critical questions.
© 2006 Elsevier B.V. All rights reserved.

Keywords: Multiple sclerosis; Longitudinal study; CLIMB

Contents

* Corresponding author. Tel.: +1 617 525 5300; fax: +1 617 525 5252.
E-mail address: hweiner@rics.bwh.harvard.edu (H.L. Weiner).

1568-9972/$ - see front matter © 2006 Elsevier B.V. All rights reserved.
doi:10.1016/j.autrev.2006.02.012

1. Introduction

Multiple sclerosis (MS), similar to other chronic autoimmune diseases, has a heterogeneous disease course. The intersection of biological and clinical variables in MS has yet to be fully understood and presents a significant challenge in the daily management of MS patients. We propose a comprehensive longitudinal study of MS integrating both clinical and biological variables, which may potentially provide a model to be applied to similar autoimmune diseases.

2. Predicting the clinical course of MS

There have been several reports based on the longitudinal follow-up of 1099 patients seen between 1972 and 1984 in London, Ontario and continued follow-up of these patients has now extended beyond 25 years. Weinshenker et al., [1] found that male gender, older age at onset, cerebellar involvement at onset, higher number of attacks in the first two years, shorter first interattack interval, and a shorter time to reach a Disability Status Score (DSS) [2] of 3 were predictors of a higher disability. Confavreux et al. further established that early clinical variables, similar to previous studies, significantly influenced the time from the onset of MS to an Expanded DSS [3] of 4, indicating a significant limitation in ambulation, but interestingly these factors did not influence subsequent disability [4].

Recently published studies from Iceland and Omstead County, indicate there may be populations of MS patients with a fairly benign course of the disease [5–7] although patients may not always remain with a benign course as demonstrated by Hawkins and McDonnell [8]. These studies demonstrate the heterogeneity of disease progression and the potential for subgroups of MS patients.

All of these studies have provided a tremendous insight into the clinical progression of MS and the clinical prognostic features of the disease that served to guide physicians caring for MS patients. However, these studies were completed prior to the era of treatment and lack MRI imaging, measurements of cognitive function as well as immunological or genetic markers, therefore they have a limited application to the current and future care of MS patients.

3. The current era of treatment in MS

The study of multiple sclerosis entered a new era with the FDA approval of beta interferon in 1993. Currently, FDA approved disease-modifying therapy for relapsing–remitting patients includes interferon beta-1b, interferon beta-1a and glatiramer acetate. All of these therapies have been widely accepted and are most commonly initiated at the time of diagnosis, although the long-term effectiveness of therapeutic interventions for MS remains largely unknown. Patients refractory to disease-modifying therapy may be treated with oral or intravenous (IV) immunosuppressant therapy. The most common IV therapies used are mitoxantrone and cyclophosphamide, both have been demonstrated to be effective at stabilizing active relapsing–remitting and early secondary progressive disease [9]. Thus, the MS paradigm has radically changed with the current treatment options available for MS that mandate the redefinition of the disease course in the current era of treatment.

4. Magnetic resonance imaging and MS

MRI-derived measures of lesion accrual and tissue loss have acquired a central role in the understanding of MS disease evolution, pathogenesis of symptoms, and prediction of clinical outcome. Characteristic lesions shown by conventional MRI as hyperintensities on T2-weighted images can detect multiple aspects of MS pathology such as edema, demyelination, remyelination, axonal loss, and gliosis but is not able to distinguish between them with a high rate of specificity [10]. Gadolinium (Gd+) enhancement is a marker of blood–brain barrier impairment that has been found to be associated with new lesions and is considered to be a marker of active inflammation [11]. Recent pathological studies have emphasized that axonal loss is a prominent feature in demyelinating lesions and is the potential cause for irreversible neurological damage and subsequent disability [12]. Providing evidence that axonal loss may correlate with progressive disability, studies of cerebral [13] and spinal cord atrophy [14] as well as magnetic resonance spectroscopy (MRS), measuring decreases in *N*-acetylaspartate (NAA) as a marker of axonal damage, [15] have all been found to correlate with disability in MS. Additionally, longitudinal MRI studies are now emerging

demonstrating the significance of MRI markers of disease activity and long-term disability [13,16].

Thus, the continued study of the use of MRI as a biological marker of disease activity in MS and the longitudinal changes associated with these various MRI markers is imperative for the fundamental understanding of disease progression and to begin to identify subgroups of MS patients.

5. Immunology and genetic studies in MS

MS is a chronic inflammatory disease of CNS myelin postulated to be a Th1 type cell-mediated autoimmune disease [17]. A Th1-type immunologic milieu exists in the disease and becomes more pronounced as the disease changes from the relapsing–remitting to the progressive stages; this milieu involves both T cells and non-T cells. Defects of regulation contribute to the disease process and involve cytokines such as IL-4, IL-10 and TGF-β and regulatory T cells. A great deal of progress has been made in understanding immune abnormalities in MS, in the past decade immune modulating therapies have been approved for MS and have become in more widespread use. Important unanswered questions central to understanding the immunology of MS not only include basic immunologic questions which seek a more precise definition of the nature of immune dysregulation that occurs in the disease, but clinically related immunologic questions that seek to define how immune abnormalities are related to disease stage, progression, MRI changes and current immunomodulatory therapy.

Numerous studies have been performed to find genes associated with multiple sclerosis [18,19]. Although there are reports of several genes that may be linked to MS, the locus that most consistently has been shown in virtually all studies to be associated with MS is the histocompatibility locus and HLA-DR2 [18,20,21]. In addition, there is evidence that the presence of the APOE-e4 allele is associated with a more severe course of MS and a recent paper reported the accelerated evolution of brain atrophy and "black holes" in MS patients with APOE-e4 [22]. With recent advances in genetics, an international consortium is currently planning to use haplotype maps to screen the entire genome to identify candidate MS genes, thus representing a rapidly expanding field which we expect will be a critical advancement to the understanding of MS disease progression [23–25].

6. Cognitive dysfunction in MS

Cognitive deficits including poor attention, decreased memory, and reduced speed of information processing occur in approximately 40% of patients with MS [26,27]. Recent evidence suggests that cognitive dysfunction in MS may begin early in the disease process. Feinstein et al., [28] demonstrated deficits in attention and working memory in 42 patients with acute optic neuritis. Patients with abnormalities on MRI were more likely to display impairments in cognitive function. In a study of 67 patients with new onset neurological symptoms and the diagnosis of probable MS, Achiron and Barak [29] reported cognitive impairment in 54% of patients. There were no correlations between cognitive impairment and MRI measures of disease burden.

Long-term studies of cognitive dysfunction in MS are limited, however Amato et al., [30] found that after ten years of follow-up cognitive dysfunction had emerged and progressed in a sizable proportion of patients compared to baseline and at four years. Additional long-term cognitive studies are required to more fully understand the precise course of cognitive dysfunction in MS.

7. CLIMB: Comprehensive Longitudinal Investigation of Multiple sclerosis at the Brigham and Women's Hospital, Partners MS Center

We have initiated a study that will monitor annual clinical and laboratory examinations on a cohort of 1000 MS patients enrolled within 3 years of diagnosis and collect data from neurological examinations, MRI imaging, immunologic, genetic, neuropsychological, and quality of life studies over the next twenty years. We currently have 515 patients enrolled in CLIMB.

7.1. Study methods

All patients ≥ 18 years of age with a definitive diagnosis of MS within the last three years will be eligible to participate. Patients may be untreated or receiving any of the standard or investigational therapies for MS.

Patients will undergo semi-annual comprehensive neurological examinations, annual psychosocial and annual cognitive evaluations. Patients will have an annual MRI scheduled within four weeks of the annual neurological examination. Patients will be imaged on a 1.5 T whole-body MRI system (GE Medical Systems). The following quantitative brain and spinal cord MRI measures will be derived: 1) T2-hyperintense lesion volume; 2) brain parenchymal fraction; 3) number of new gadolinium-enhancing lesions; and 4) total number of gadolinium-enhancing lesions.

At enrollment, patients will be tissue typed since tissue type may modify the immune response. Intracytoplasmic staining will be used to characterize the cells that preferentially express a particular cytokine. Intracytoplasmic staining will be performed for T cells (IL-4, IL-5, TGF-β, TNF-α, and IFN-γ) and monocytes (IL-10, IL-12, IL-23, TNF, and TGF-β). Surface staining will be used to measure the surface expression of activation molecules on CD4+ T cells and monocytes. CD25, CD26, Class II MHC expression on T cells, and CD40, CD80 and CD86 expression on monocytes will be examined. CD40, CD80 and CD86 expression on monocytes will be studied to establish the role of expression of costimulatory molecules and the CD40–CD40L pathway. A serum sample will be collected from each patient. Serum levels of IL-12, IFN-γ, ICAM, and TNF-α will be measured by ELISA. RNA will be extracted and gene product expression for IL-23, IL-12, IFN-γ, TNF, IL-10 and TGF-β will be measured. Testing for APOE-e4 allele will be completed based on its reported association with accelerated evolution of brain atrophy [22].

7.2. Statistical methods

Mixed effects regression models will be used to assess the cross-sectional associations between clinical, psychosocial and immunologic variables, within and between visit times, while controlling for possible confounders (e.g., time since diagnosis, age at diagnosis, gender, HLA). Specifically, EDSS will be our clinical outcome and we will calculate its association with MRI metrics and immunologic variables. We will adjust for repeated observations of subjects through the use of random effects in the models [31]. Multivariate survival models will be used to determine predictors of disease course.

In addition to long-term progression, we will determine the factors associated with short-term progression by fitting Markov transitional models to our data. These models offer a novel analytic tool for MS progression that exploits the ordinal nature of the EDSS. They regress the transitions of patients from one level of EDSS to another in subsequent visits on selected covariates [31,32]. We will include random effects in our model to account for unmeasured covariates. Once the model is fitted, a transition probability matrix can be generated for each subject (according to his/her covariates), and a probability of progression over time can be calculated and drawn. This provides formal as well as informal graphical tools to compare group of subjects.

8. Conclusions

There are many applications of the CLIMB study; most importantly we will attempt to define the outcome of the disease given its early presentation, which will enable clinicians to counsel patients about likely prognosis. It is clear that MS is a heterogeneous disease, but subcategories are not well understood or defined. The identification of MS subgroups may demonstrate immunologic, pathologic, and genetic differences, as well as differences in their response to drug treatment and will contribute significantly to the understanding of the disease pathogenesis. We expect the knowledge gained through the CLIMB study will enable clinicians to provide better clinical care to MS patients and to contribute to the advancement of MS research. In conclusion, the study of all chronic autoimmune diseases is inherently complicated, however as the advancement of biological markers continues, the CLIMB study may provide a model for the investigation of autoimmune diseases similar to MS.

Take-home messages

- Advancements in treatment, MRI imaging, immunology and genetics has created a platform for the comprehensive study of MS.
- A longitudinal study of multiple sclerosis incorporating both clinical and biological markers may serve as a paradigm for the investigation of other chronic autoimmune diseases.

References

[1] Weinshenker BG, Rice GP, Noseworthy JH, Carriere W, Baskerville J, Ebers GC. The natural history of multiple sclerosis: a geographically based study. 3. Multivariate analysis of predictive factors and models of outcome. Brain 1991;114 (Pt 2):1045–56.

[2] Kurtzke JF, Beebe GW, Nagler B, Nefzger MD, Auth TL, Kurland LT. Studies on the natural history of multiple sclerosis. V. Long-term survival in young men. Arch Neurol 1970;22:215–25.

[3] Kurtzke J. Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). Neurology 1983;33: 227–31.

[4] Confavreux C, Vukusic S, PA. Early clinical predictors and progression of irreversible disability in multiple sclerosis: an amnesic process. Brain 2003;126:770–82.

[5] Pittock SJ, Mayr WT, McClelland RL, al e. Disability profile of MS did not change over 10 years in a population-based prevalence cohort. Neurology 2004;62:601–6.

[6] Benedikz J, al e. The natural history of untreated multiple sclerosis in Iceland. A total population-based 50 year prospective study. Clin Neurol Neurosurg 2002;104:208–10.

[7] Pittock SJ, McClelland RL, Mayr WT, Jorgensen NW, Weinshenker BG, Noseworthy J, et al. Clinical implications of benign multiple sclerosis: a 20-yr population-based follow-up study. Ann Neurol 2004;56:303–6.
[8] Hawkins SA, McDonnell GV. Benign multiple sclerosis? Clinical course, long-term follow up, and assessment of prognostic factors. J Neurol Neurosurg Psychiatry 1999;67:148–52.
[9] Gauthier SA, Buckle GJ, Weiner HL. Immunosuppressive therapy for multiple sclerosis. Neurol Clin 2005:247–72.
[10] Bruck W, Bitsch A, Kolenda H, Bruck Y, Stiefel M, Lassmann H. Inflammatory central nervous system demyelination: correlation of magnetic resonance imaging findings with lesion pathology. Ann Neurol 1997;42:783–93.
[11] Miller DH, Rudge P, Johnson G, Kendall BE, Macmanus DG, Moseley IF, et al. Serial gadolinium enhanced magnetic resonance imaging in multiple sclerosis. Brain 1988;111(Pt 4):927–39.
[12] Trapp B, Peterson J, Ransohoff R, Rudick R, Mork S, Bo L. Axonal transection in the lesions of multiple sclerosis. N Engl J Med 1998;338:278–85.
[13] Fisher E, Rudick RA, Simon JH, Cutter G, Baier M, Lee JC, et al. Eight-year follow-up study of brain atrophy in patients with MS. Neurology 2002;59:1412–20.
[14] Stevenson VL, Leary SM, Losseff NA, Parker GJ, Barker GJ, Husmani Y, et al. Spinal cord atrophy and disability in MS: a longitudinal study. Neurology 1998;51:234–8.
[15] Fu L, Matthews PM, De Stefano N, Worsley KJ, Narayanan S, Francis GS, et al. Imaging axonal damage of normal-appearing white matter in multiple sclerosis. Brain 1998;121(Pt 1):103–13.
[16] Brex POC, O'Riordan JI, M.S., Thompson AJ, Miller DH. A longitudinal study of abnormalities on MRI and disability from multiple sclerosis. N Engl J Med 2002;346:158–63.
[17] Hafler DA, Weiner HL. Immunologic mechanisms and therapy in multiple sclerosis. Immunol Rev 1995;144:75–107.
[18] Kantarci OH, de Andrade M, Weinshenker BG. Identifying disease modifying genes in multiple sclerosis. J Neuroimmunol 2002;123:144–59.
[19] Kenealy SJ, Pericak-Vance MA, Haines JL. The genetic epidemiology of multiple sclerosis. J Neuroimmunol 2003;143: 7–12.
[20] Hauser SL, Fleischnick E, Weiner HL, Marcus D, Awdeh Z, Yunis EJ, et al. Extended major histocompatibility complex haplotypes in patients with multiple sclerosis. Neurology 1989;39:275–7.
[21] GAMES. A meta-analysis of whole genome linkage screens in multiple sclerosis. J Neuroimmunol 2003;143:39–46.
[22] Enzinger C, Ropele S, Smith S, Strasser-Fuchs S, Poltrum B, Schmidt H, et al. Accelerated evolution of brain atrophy and "black holes" in MS patients with APOE-epsilon 4. Ann Neurol 2004;55:563–9.
[23] Daly MJ, Rioux JD, Schaffner SF, Hudson TJ, Lander ES. High-resolution haplotype structure in the human genome. Nat Genet 2001;29:229–32.
[24] Gabriel SB, Schaffner SF, Nguyen H, Moore JM, Roy J, Blumenstiel B, et al. The structure of haplotype blocks in the human genome. Science 2002;296:2225–9.
[25] The International HapMap Project. Nature 2003;426:789–96.
[26] Rao SM, Leo GJ, Bernardin L, Unverzagt F. Cognitive dysfunction in multiple sclerosis. I. Frequency, patterns, and prediction. Neurology 1991;41:685–91.
[27] McIntosh-Michaelis SA, Roberts MH, Wilkinson SM, Diamond ID, McLellan DL, Martin JP, et al. The prevalence of cognitive impairment in a community survey of multiple sclerosis. Br J Clin Psychol 1991;30(Pt 4):333–48.
[28] Feinstein A, Youl B, Ron M. Acute optic neuritis. A cognitive and magnetic resonance imaging study. Brain 1992;115(Pt 5):1403–15.
[29] Achiron A, Barak Y. Cognitive impairment in probable multiple sclerosis. J Neurol Neurosurg Psychiatry 2003;74:443–6.
[30] Amato MP, Ponziani G, Siracusa G, Sorbi S. Cognitive dysfunction in early-onset multiple sclerosis: a reappraisal after 10 years. Arch Neurol 2001;58:1602–6.
[31] Diggle PH, Heagerty PJ, Liang K-Y, Zeger S. Analysis of longitudinal data. Second ed. New York: Oxford University Press; 2002.
[32] Albert PS. A Markov model for sequences of ordinal data from a relapsing–remitting disease. Biometrics 1994;50:51–60.

IgG-antiphospholipid antibodies in follicular fluid of IVF-ET patients are related to low fertilization rate of their oocytes

Patients undergoing in vitro fertilization and embryo transfer (IVF-ET) failures show an increased incidence of antiphospholipid antibodies (aPL) in their blood. The physiological manifestations of aPL in this patient group are nonetheless been documented. Here, Matsubayashi H. et. al. (Am J Reprod Immunol 2006; 55: 341-8) questioned whether aPL if found in follicular fluids (FFs) could result in embryonic damage. Blood from 44 patients with three or more IVF-ET failures were tested for the presence of immunoglobulin (Ig)G, IgM and IgA aPL. Both the 29 aPL-positive and 15 aPL-negative patients gave permission, for FF collection during their next IVF-ET attempt for additional aPL determinations. Patients with no aPL in their blood, had no aPL in their FFs. Patients with IgG and/or IgM aPL in their blood had IgG but not IgM in their respective FFs. The presence of IgG aPL in FFs and increased infertility length were significantly related to lower fertilization rates, independently. Follicular fluid IgG aPL appears as a risk factor in association with successful IVF-ET outcomes.