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Can Virus Infections Trigger Autoimmune Disease?

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Introduction

This review will examine various infectious parameters that could lead to enhancement or exacerbation of autoimmune disease. Multiple sclerosis (MS) will be the prototype human autoimmune disease detailed and experiments relating to the pathogenesis of MS will use data derived from an experimental animal model, experimental allergic encephalomyelitis (EAE). Other autoimmune diseases will be included for comparison.

MS is the most common human demyelinating disease with a prevalence rate between 50–100 per 100,000 Caucasians [1]. Other ethnic groups have lower but significant prevalence rates. Women are more afflicted than men by more that a 2:1 ratio. The inflammatory demyelinating lesions are limited to the central nervous system (CNS) [2]. In most cases, oligoclonal IgG bands are present in the cerebral spinal fluid and a mild mononuclear pleocytosis may be present. Clinical features include vision loss due to optic neuritis, weakness of the limbs and sensory disturbances with some memory and cognitive impairment. The clinical course can include relapses and remissions and/or a progressive course. MS is immune mediated and, while not conclusively shown, thought to be caused by autoreactive myelin-specific Th1 $CD4^+$ T cells [3].

Genetic Involvement in MS

MS has been shown to have a genetic component. This concept comes from two lines of investigation. First, in studies with monozygotic twins, if one twin developed MS there was about a 25–30% chance the other twin would also develop MS [4–6]. Second, there is a HLA association in MS patients with HLA DRw15,DQw6,Dw2 in Caucasian Europeans and North Americans [7, 8] and a secondary association with DRw15,DQw6,Dw2 in Swedish MS patients [9].

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These sorts of data indicate that while there is an important genetic component to MS, environmental factors such as infections play an important role in the pathogenesis.

Interestingly when twins are studied in the context of infectious disease, concordance rates for highly infectious diseases, such as measles virus infection, are greater than 90% for monozygotic twins. However, for other infectious diseases such as pneumonia, concordance rates are in the range of 30%, approximating that for MS [10].

Evidence for Environmental Factors

Further evidence supporting an infectious contribution to the pathogenesis of MS comes from epidemiological studies. Epidemiological data indicate that there is a latitude distribution (North/South in the Northern Hemisphere) in the incidence of MS. The northern latitudes in Europe have a higher incidence of MS. Similarly, there is an increased incidence of MS in northern part of the United States (above the 37th parallel). Further when individual countries were surveyed for the incidence of MS, a clustering of MS cases has been reported [1]. This clustering was also seen when the same regions were resurveyed a generation later. These studies led Kurtzke to suggest, 'the occurrence of MS is intrinsically related to geography, and therefore MS can be defined as an acquired, exogenous, environmental disease' [1]. It appears that MS is a place-related disorder. The areas of the world most affected are Northern Europe or areas colonized by the Europeans [11]. These regions include Canada, the United States, Australia, New Zealand and South Africa. Kurtzke speculated that the spread of MS from Scandinavia to other regions was too rapid to be due strictly to genes or genetics, but that an environmental agent was most likely the cause [1].

From migration studies, immigrants tended to retain the MS risk of their birthplace. Migrants from high-risk areas moving to low-risk regions usually retained their high-risk phenotype [12]. However,

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age must be a consideration in the MS migrant studies [13–18]. An individual who moves prior to the age of 15 will acquire the risk associated with the area to which she/he moves; whereas if migration occurs after the age of 15, the individual keeps the risk associated with the area in which they were born. This observation could relate to the number and types of infections in a particular region that an individual is exposed to and when in life they were exposed. These data are consistent with the hypothesis that infections early in life could subclinically prime genetically susceptible individuals for MS. Interestingly this may also be the case for insulin dependent diabetes mellitus (IDDM) (reviewed in [19]).

Several studies provide interesting data documenting the infectious nature of MS. Studies of Iceland indicate that there was an increase in the incidence of MS following World Wars I and II when British, Canadian and American soldiers occupied Iceland [20]. Similarly, in five surveys by four groups studying the Shetland-Orkney Islands, there was a marked increase in MS following the Second World War [21–24]. Epidemiological surveys of the Faroe Islands also demonstrated an increase in MS following the Second World War and the locations of British troop encampments strongly correlated with the place of residence of all MS patients [25–27]. MS did not appear to spread to surrounding areas that did not house British troops. In addition, there were few, if any, documented cases of MS prior to 1943 in the Faroe Islands.

Infections Associated with Exacerbations

Infections have long been associated with attacks of MS. Rapp *et al*. [28] found an increase in MS exacerbations with patients experiencing bacterial infections. Metz *et al*. [29] have demonstrated that recurrent urinary tract infections were associated with acute exacerbations and neurologic progression. More recent studies [30–32] found that there was an association between upper respiratory viral infections and exacerbations of MS. These studies confirm earlier work by Sibley *et al*. [33] who studies 170 patients with MS over an 8-year period. These investigators concluded that viral-like infections were temporally associated with exacerbations of MS. In all of these studies, no common infection, either bacterial or viral was found, but rather infections were frequently found in temporal association with clinical attacks of MS. There is an apparent paradox relating to infection. Well over 24 viral agents have been isolated from MS patients [34]. However, no virus has been identified as the 'MS virus', yet viral infections are often seen in temporal association with exacerbations [32, 33].

This is in contrast to individuals who acquire a congenital rubella infection. These individuals have a much greater incidence of diabetes [35, 36]. Autoantibodies to islet cells were found in greater than 20% of the total population with congenital rubella syndrome [37]. This study also investigated the HLA association [37]. They concluded that the same genetic and immunologic features seen in classic IDDM, such as the presence of HLA DR3 and the absence of DR2 and the occurrence of autoantibodies, was also seen in those IDDM patients congenitally infected with rubella. Thus, here there is a clear association with rubella virus infection and IDDM. IDDM due to congenital rubella has declined owing to the successful vaccination campaign against mumps.

Experimental Allergic Encephalomyelitis (EAE) as a Model for MS

EAE is a widely used animal model for MS. It can be induced in a variety of vertebrate species using spinal cord homogenates, myelin, and proteins comprising myelin, such as myelin basic protein (MBP), myelin proteolipid protein (PLP), myelin associated glycoprotein (MAG), myelin oligodendrocyte glycoprotein (MOG), in adjuvants such as complete Freund's adjuvant (CFA). MHC-restricted class II peptides (encephalitogenic peptides) have been identified and can also be used to induce EAE. Depending on the species and/or strain, different clinical forms of EAE can be seen. For example, Lewis rats sensitized with spinal cord homogenate in CFA develop an acute attack of EAE with few if any relapses. In contrast, SJL/J mice sensitized with $PLP_{139–151}$ in CFA undergo a relapsing-remitting disease course, whereas AS.W mice die of a progressive disease [38]. Clinical signs include weight loss, ataxia and incontinence with flaccid or spastic hind limb paralysis.

CNS lesions are inflammatory in nature and often include demyelination. These lesions are similar to early acute lesions seen in MS. However, the distribution of lesions is somewhat different in EAE where spinal cord lesions are more prominent, whereas in MS brain lesions are more common. With time large plaque-like lesions are present in MS. The only model of EAE that approximates plaque-like lesions is MOG induced EAE (Figure 1). Interestingly in MOG induced EAE, antibodies play a prominent role in the disease whereas other models of EAE are thought to be strictly a Th1 $CD4^+$ T cell-mediated disease. In most models of EAE, myelin-specific Th1 $CD4^+$ T cells can adoptively transfer EAE to naïve recipients. In MS it is thought that these cells are also important in the pathogenesis.

Vaccination and MS/EAE

To date, no immunization or vaccination has been demonstrated to induce exacerbations in MS. Four studies conducted in the mid-1970s to the mid-1980s found no association or 'cause and effect' relationship between swine influenza vaccine and MS [39–42]. This was a major concern since the swine influenza vaccine may have had an increase in the relative risk for

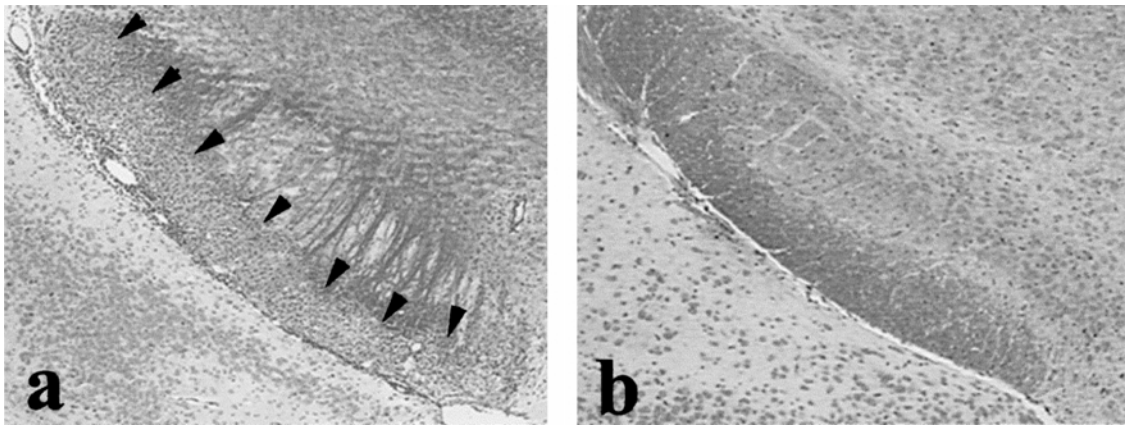

Figure 1. Plaque-like demyelination in primary progressive experimental allergic encephalomyelitis (PP-EAE) induced with myelin oligodendrocyte glycoprotein (MOG). (A) Almost entire cerebral peduncle was demyelinated in SJL/J mouse with $MOG_{92–106}$-induced-PP-EAE. (B) Normal cerebral peduncle in control mouse. Luxol fast blue stain, magnification×40.

Guillain-Barre syndrome within the first 6 weeks after vaccination [43]. More recently several studies have shown that there were no increases in exacerbations of MS in influenza virus vaccinated individuals [44–47]. Interestingly in one study, De Keyser *et al*. [45] found that in a group of 180 patients with relapsing-remitting MS an exacerbation occurred within the following 6 weeks in 33% after influenza illness. The exacerbation rate in influenza vaccinated patients was only 5%. They concluded that annual influenza vaccination should be offered routinely to all patients with relapsing MS. Recently there has been some controversy over the hepatitis B virus vaccine and MS. Investigators reviewing the evidence have concluded that there is no association between hepatitis B virus vaccination and increased risk for MS [48–50].

On the other side of the coin, several groups have attempted to use a variety of schemes to vaccinate against autoimmune disease. Some of these will be discussed below in the context of EAE.

Since the time of Jenner, cowpox and vaccinia viruses were used to vaccinate humans against smallpox. More recently, vaccinia virus has been used to express proteins during viral infection, including self proteins. This approach was used to modulate EAE. Recombinant vaccinia virus encoding PLP (VV_{PLP}) was used to infect mice. These 'vaccinated' mice were then challenged with $PLP_{139–151}$ in CFA and followed for EAE [51]. We observed that the first acute attack of EAE was exacerbated versus control mice vaccinated with VV_{SC11}, a recombinant virus encoding β-galactosidase, or in mice not vaccinate and challenged with $PLP_{139–151}$ in CFA. Interestingly, we found that mice vaccinated with VV_{PLP} and challenged with $PLP_{139–151}$, after the first initial exacerbated attack, seldom had a relapse; and, if it did occur, the relapse was very mild [52]. This was in contrast to mice infected with control virus VV_{SC11} or mock infected and challenged with $PLP_{139–151}$ where they displayed a relapsing-remitting disease course. Here virus encoding a cross-reacting determinant (molecular mimicry) was able to exacerbate the initial attack of EAE but limit subsequent attacks.

In a different system, mice infected with a recombinant vaccinia virus encoding an encephalitogenic epitope $MBP_{1–23}$, ($VV_{MBP1–23}$) and challenged with $MBP_{1–20}$ or the intact MBP molecule in CFA were protected from the development of clinical signs or pathologic changes of EAE [53]; however, we found that mice were not protected when challenged with spinal cord homogenate. When immunologic parameters were measured, proliferative responses to MBP were decreased as well as delayed type hypersensitivity (ear swelling) responses associated with $CD4^+$ Th1 T cells. T cells from $VV_{MBP1–23}$ vaccinated mice were not able to adoptively transfer EAE to naïve mice. Using a similar approach in marmosets, Genain *et al*. [54] confirmed the above mouse experiments. Here marmosets were infected with a recombinant vaccinia virus encoding intact human MBP and challenged with whole white matter in CFA and *Bordetela pertussis* 3–5 weeks after the last vaccination. Some of the monkeys received more than one vaccination. They found amelioration of disease in some of the animals. Therefore, depending on the CNS protein encoded by the recombinant vaccinia virus either an enhancement or protection is observed. It may be that depending on what epitopes a virus may have in common or similar with self molecules could dictate the response to subsequent challenge.

DNA Vaccination as a Means to Modulate CNS Autoimmune Disease

We initiated studies to determine if immunization with plasmid DNA encoding PLP could prime PLP-specific immune responses. Several possibilities could arise from such a scheme. First, inoculation with a cDNA encoding PLP could suppress EAE by inducing MHC class I restricted $CD8^+$ suppressor T cells, since

in some models of EAE $CD8^+$ T cells have been shown to modulate EAE [55]. Second, anergy or tolerance could arise due to expression of PLP in non-professional antigen presenting cells (APCs). Muscle cells could present peptide without appropriate co-stimulation [56]. Third, inoculation may induce EAE or enhance subsequent induction of EAE, since cDNA inoculation has been shown to induce Th1-like immune responses [57]. Fourth, cDNA vaccination encoding PLP could produce a different CNS disease through a PLP specific $CD8^+$ T cell response.

Using cDNAs encoding PLP or encephalitogenic PLP peptides, $PLP_{139–151}$ and $PLP_{178–191}$, we tested whether vaccination could modulate the development of relapsing-remitting EAE using PLP peptides in CFA. When mice were vaccinated with the cDNA without subsequent peptide challenge, proliferative responses were detected to PLP but no inflammatory lesions were present in the CNS. Upon challenge of cDNA vaccinated mice with PLP peptides a more severe EAE was seen both clinically and histologically versus control mice. Measuring anti-PLP isotype antibody responses indicated that a Th1 response was favoured. Interestingly, Ruiz *et al*., using a very similar system, came to the opposite conclusion [58]. They found amelioration of clinical signs of EAE in their system. A reduction in Th1 type cytokine mRNAs was also noted in the CNS. Lobell *et al*. [59] found that vaccination of Lewis rats with a cDNA encoding a Lewis rat encephalitogenic MBP peptide only suppressed disease when the peptide was targeted to Fc receptors using an analogue of protein A. Targeting the peptide to Fc was essential for the modulation [59]. Protection was encephalitogenic epitope specific and did not appear to involve bystander suppression [60]. We found that EAE as well as Theiler's virus infection of mice (a viral model for MS) could be exacerbated by injection of mice with just plasmid DNA. The extent of enhancement correlated with the number of injections of plasmid DNA. Increases in proinflammatory cytokines were observed [61]. Segal *et al*. [62] showed similar results demonstrating that CpG containing oligonucleotides could replace mycobacteria in CFA for active sensitization. The enhancement was dependent on IL-12 [63]. It still remains to be determined whether this will be a viable means to down-modulate EAE and/or if this technique can be applied to MS and other autoimmune disorders.

Virus Infection having Molecular Mimicry can Subclinically Prime for Autoimmune Disease that can be Exacerbated by a Non-related Infection

We have recently used cDNA encoding PLP to subclinically prime animals for autoimmune CNS disease. Young SJL/J mice were inoculated three times with a cDNA encoding PLP. Mice developed neither clinical nor inflammatory lesions in the CNS even though lymphoid cells from these mice proliferated in response to PLP encephalitogenic peptides. When these mice were given a non-specific immunologic challenge in the form of CFA (a potent IL-12 inducer) mice developed inflammatory lesions in the white matter of the CNS. Instead of challenging mice with CFA, infection of mice with a recombinant vaccinia virus encoding β-galactosidase could replace CFA in inducing inflammatory lesions in white matter in the CNS. Interestingly, infection of mice with recombinant vaccinia viruses encoding PLP, myelin associated glycoprotein or glial fibrillary acidic protein could replace cDNA in that after infection, when mice were challenged with CFA, about 90% of mice develop inflammatory lesions.

Summary

We have attempted to provide a review of information concerning infection and vaccination relating to autoimmune disease. In the limited amount of space, not all modalities were covered but important features summarized. What is emerging is that natural infections can cause exacerbations of autoimmune disease. This is most likely due to the induction of IL-12, IL-6 and IFN-γ. To date there are no data demonstrating an association between vaccination and induction of autoimmunity or exacerbations. Interestingly in some instances vaccination appears to lower the risk of exacerbations. However, in the future it will be of interest to review the data about DNA vaccines and their use to prevent infections and alter autoimmunity.

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References

1. Kurtzke J.F. 1997. The Epidemiology of Multiple Sclerosis. In *Multiple Sclerosis: Clinical and Pathogenetic Basis* C.S. Raine, H.F. McFarland, W.W. Tourtellotte, eds. Chapman & Hall, London, pp. 91–140
2. Dejong R.N. 1970. Multiple sclerosis. History, definition and general considerations. In *Multiple Sclerosis and Other Demyelinating Diseases* P.J. Vinken, G.W. Bruyn, eds. North-Holland Publishing Company, Amsterdam, pp. 45–62
3. Noseworthy J.H., Lucchinetti C., Rodriquez M., Weinshenker B.G. 2000. Multiple sclerosis. *N. Engl. J. Med.* **343:** 938–952
4. Ebers G.C., Bulman D.E., Sadovnick A.D., Paty D.W., Warren S., Hader W., Murray T.J., Seland T.P., Duquette P., Grey T., Nelson R., Nicolle M., Brunet D. 1986. A population-based study of multiple sclerosis in twins. *N. Engl. J. Med.* **315:** 1638–1642
5. Sadovnick A.D., Armstrong H., Rice G.P.A., Bulman D., Hashimoto L., Paty D.W., Hashimoto S.A., Warren S., Hader W., Murray T.J., Seland T.P., Metz L., Bell R.,

Duquette P., Gray T., Nelson R., Weinshenker B., Brunet D., Ebers G.C. 1993. A population-based study of multiple sclerosis in twins: Update. *Ann. Neurol.* **33:** 281–185

6. Mumford C.J., Wood N.W., Kellar-Wood H., Thorpe J.W., Miller D.H., Compston D.A. 1994. The British Isles survey of multiple sclerosis in twins. *Neurology* **44:** 11–15
7. Kalman B., Lublin F.D. 1999. The genetics of multiple sclerosis. A review. *Biomed. Pharmacother.* **53:** 358–370
8. Martin R. 1997. Genetics of multiple sclerosis—how could disease-associated HLA-types contribute to pathogenesis? *J. Neural. Transm. Suppl.* **49:** 177–194
9. Olerup O., Hillert J. 1991. HLA class II-associated genetic susceptibility in multiple sclerosis: a critical evaluation. *Tissue Antigens* **38:** 1–15
10. Vogel F., Motulsky A.G. 1997. *Human Genetics: Problems and Approaches*. Springer, Berlin and New York
11. Poser C.M. 1994. The dissemination of multiple sclerosis: A Viking saga? A historical essay. *Ann. Neurol.* **36**(Suppl 2): S231–S243
12. Kurtzke J.F. 1993. Epidemiologic evidence for multiple sclerosis as an infection. *Clin. Microbiol. Rev.* **6:** 382–427
13. Alter M., Leibowitz U., Speer J. 1966. Risk of multiple sclerosis related to age at immigration to Israel. *Arch. Neurol.* **15:** 234–237
14. Kurtzke J.F., Dean G., Botha D.P. 1970. A method for estimating the age at immigration of white immigrants to South Africa, with an example of its importance. *S. Afr. Med. J.* **44:** 663–669
15. Kurtzke J.F., Kurland K.T., Goldberg I.D. 1971. Mortality and migration in multiple sclerosis. *Neurology* **21:** 1186–1197
16. Dean G., Kurtzke J.F. 1971. On the risk of multiple sclerosis according to age at immigration to South Africa. *Br. Med. J.* **3:** 725–729
17. Alter M., Kahana E., Loewenson R. 1978. Migration and risk of multiple sclerosis. *Neurology* **28:** 1089–1093
18. Alter M., Okihiro M. 1971. When is multiple sclerosis acquired? *Neurology* **21:** 1030–1036
19. Leslie R.D., Elliott R.B. 1994. Early environmental events as a cause of IDDM. Evidence and implications. *Diabetes* **43:** 843–850
20. Kurtzke J.F., Gudmundsson K.R., Bergmann S. 1982. Multiple sclerosis in Iceland: 1. Evidence of a postwar epidemic. *Neurology* **32:** 143–150
21. Sutherland J.M. 1956. Observations on the prevalence of multiple sclerosis in Northern Scotland. *Brain* **79:** 635–654
22. Allison R.S. 1963. Some neurological aspects of medical geography. *Proc. R. Soc. Med.* **56:** 71–76
23. Fog M., Hyllested K. 1966. Prevalence of disseminated sclerosis in the Faroes, the Orkneys and Shetland. *Acta. Neurol. Scand.* **42**(Suppl. 19): 9–11
24. Poskanzer D.C., Prenney L.B., Sheridan J.L., Kondy J.Y. 1980. Multiple sclerosis in the Orkney and Shetland Islands. I: Epidemiology, clinical factors, and methodology. *J. Epidemiol. Community Health* **34:** 229–239
25. Kurtzke J.F., Hyllested K., Heltberg A., Olsen A. 1993. Multiple sclerosis in the Faroe Islands. 5. The occurrence of the fourth epidemic as validation of transmission. *Acta. Neurol. Scand.* **88:** 161–173
26. Kurtzke J.F., Hyllested K. 1987. Multiple sclerosis in the Faroe Islands. III. An alternative assessment of the three epidemics. *Acta. Neurol. Scand.* **76:** 317–339
27. Kurtzke J.F., Hyllested K. 1986. Multiple sclerosis in the Faroe Islands. II. Clinical update, transmission, and the nature of MS. *Neurology* **36:** 307–328
28. Rapp N.S., Gilroy J., Lerner A.M. 1995. Role of bacterial infection in exacerbation of multiple sclerosis. *Am. J. Phys. Med. Rehabil.* **74:** 415–418
29. Metz L.M., McGuinness S.D., Harris C. 1998. Urinary tract infections may trigger relapse in multiple sclerosis. *Axone* **19:** 67–70
30. Andersen O., Lygner P.-E., Bergström T., Andersson M., Vahlne A. 1993. Viral infections trigger multiple sclerosis relapses: a prospective seroepidemiological study. *J. Neurol.* **240:** 417–422
31. Edwards S., Zvartau M., Clarke H., Irving W., Blumhardt L.D. 1998. Clinical relapses and disease activity on magnetic resonance imaging associated with viral upper respiratory tract infections in multiple sclerosis. *J. Neurol. Neurosurg. Psychiatry* **64:** 736–741
32. Panitch H.S. 1994. Influence of infection on exacerbations of multiple sclerosis. *Ann. Neurol.* **36**(Suppl): S25–S28
33. Sibley W.A., Bamford C.R., Clark K. 1985. Clinical viral infections and multiple sclerosis. *Lancet* **1:** 1313–1315
34. Johnson R.T. 1998. Chronic inflammatory and demyelinating diseases. In *Vital Infections of the Nervous System*. Lippincott-Raven, Philadelphia and New York, pp. 227–264
35. Forrest J.M., Menser M.A., Burgess J.A. 1971. High frequency of diabetes mellitus in young adults with congenital rubella. *Lancet* **7720:** 332–334
36. Smithells R., Sheppard S., Marshall W., Peckham C. 1978. Congenital rubella and diabetes mellitus. *Lancet* **i:** 439
37. Ginsberg-Fellner F., Witt M.E., Fedun B., Taub F., Doberson M.J., McEvoy R.C., Cooper L.Z., Notkins A.L., Rubinstein P. 1985. Diabetes mellitus and autoimmunity in patients with the congenital rubella syndrome. *Rev. Infect. Dis.* **7**(Suppl 1): S170–S176
38. Tsunoda I., Kuang L.-Q., Theil D.J., Fujinami R.S. 2000. Antibody association with a novel model for primary progressive multiple sclerosis: Induction of relapsing-remitting and progressive forms of EAE in $H2^s$ mouse strains. *Brain Pathol.* **10:** 402–418
39. Sibley W.A., Bamford C.R., Laguna J.F. 1976. Influenza vaccination in patients with multiple sclerosis. *JAMA.* **236:** 1965–1966
40. Myers L.W., Ellison G.W., Lucia M., Novom S., Holevoet M., Madden D., Sever J., Noble G.R. 1977. Swine influenza virus vaccination in patients with multiple sclerosis. *J. Infect. Dis.* **136**(Suppl): S546–S554
41. Kurland L.T., Molgaard C.A., Kurland E.M., Erdtman F.J., Stebbing G.E.T. 1984. Lack of association of Swine flu vaccine and rheumatoid arthritis. *Mayo Clin. Proc.* **59:** 816–812
42. Bamford C.R., Sibley W.A., Laguna J.F. 1978. Swine influenza vaccination in patients with multiple sclerosis. *Arch. Neurol.* **35:** 242–243
43. Safranek T.J., Lawrence D.N., Kurland L.T., Culver D.H., Wiederholt W.C., Hayner N.S., Osterholm M.T., O'Brien P., Hughes J.M. 1991. Reassessment of the association between Guillain-Barre syndrome and receipt of swine influenza vaccine in 1976–1977: results

of a two-state study. Expert Neurology Group. *Am. J. Epidemiol.* **133:** 940–951

44. Mokhtarian F., Shirazian D., Morgante L., Miller A., Grob D., Lichstein E. 1997. Influenza virus vaccination of patients with multiple sclerosis. *Mult. Scler.* **3:** 243–247
45. De Keyser J., Zwanikken C., Boon M. 1998. Effects of influenza vaccination and influenza illness on exacerbations in multiple sclerosis. *J. Neurol. Sci.* **159:** 51–53
46. Salvetti M., Pisani A., Bastianello S., Millefiorini E., Buttinelli C., Pozzilli C. 1995. Clinical and MRI assessment of disease activity in patients with multiple sclerosis after influenza vaccination. *J. Neurol.* **242:** 143–146
47. Miller A.E., Morgante L.A., Buchwald L.Y., Nutile S.M., Coyle P.K., Krupp L.B., Doscher C.A., Lublin F.D., Knobler R.L., Trantas F., Kelley L., Smith C.R., La Rocca N., Lopez S. 1997. A multicenter, randomized, double-blind, placebo-controlled trial of influenza immunization in multiple sclerosis [see comments]. *Neurology* **48:** 312–314
48. Shoenfeld Y., Aron M.A. 2000. Vaccination and autoimmunity-'vaccinosis': a dangerous liaison? *J. Autoimmun.* **14:** 1–10
49. Sadovnick A.D., Scheifele D.W. 2000. School-based hepatitis B vaccination programme and adolescent multiple sclerosis [letter]. *Lancet* **355:** 549–550
50. Monteyne P., Andre F.E. 2000. Is there a causal link between hepatitis B vaccination and multiple sclerosis? *Vaccine* **18:** 1994–2001
51. Barnett L.A., Whitton J.L., Wada Y., Fujinami R.S. 1993. Enhancement of autoimmune disease using recombinant vaccinia virus encoding myelin proteolipid protein [published erratum appears in *J. Neuroimmunol.* 1993:48:120]. *J. Neuroimmunol.* **44:** 15–25
52. Wang L.-Y., Theil D.J., Whitton J.L., Fujinami R.S. 1999. Infection with a recombinant vaccinia virus encoding myelin proteolipid protein causes suppression of chronic relapsing-remitting experimental allergic encephalomyelitis. *J. Neuroimmunol.* **96:** 148–157
53. Barnett L.A., Whitton J.L., Wang L.Y., Fujinami R.S. 1996. Virus encoding an encephalitogenic peptide protects mice from experimental allergic encephalomyelitis. *J. Neuroimmunol.* **64:** 163–173
54. Genain C.P., Gritz L., Joshi N., Panicali D., Davis R.L., Whitaker J.N., Letvin N.L., Hauser S.L. 1997. Inhibition of allergic encephalomyelitis in marmosets by vaccination with recombinant vaccinia virus encoding for myelin basic protein. *J. Neuroimmunol.* **79:** 119–128
55. Tsunoda I., Fujinami R.S. 1996. Two models for multiple sclerosis: Experimental allergic encephalomyelitis and Theiler's murine encephalomyelitis virus. *J. Neuropathol. Exp. Neurol.* **55:** 673–686
56. Warrens A.N., Zhang J.-Y., Sidhu S., Watt D.J., Lombardi G., Sewry C.A., Lechler R.I. 1994. Myoblasts fail to stimulate T cells but induce tolerance. *Int. Immunol.* **6:** 847–853
57. Raz E., Tighe H., Sato Y., Corr M., Dudler J.A., Roman M., Swain S.L., Spiegelberg H.L., Carson D.A. 1996. Preferential induction of a Th_1 immune response and inhibition of specific IgE antibody formation by plasmid DNA immunization. *Proc. Natl. Acad. Sci. USA.* **93:** 5141–5145
58. Ruiz P.J., Garren H., Ruiz I.U., Hirschberg D.L., Nguyen L.-V.T., Karpuj M.V., Cooper M.T., Mitchell D.J., Fathman C.G., Steinman L. 1999. Suppressive immunization with DNA encoding a self-peptide prevents autoimmune disease: modulation of T cell costimulation. *J. Immunol.* **162:** 3336–3341
59. Lobell A., Weissert R., Storch M.K., Svanholm C., de Graaf K.L., Lassmann H., Andersson R., Olsson T., Wigzell H. 1998. Vaccination with DNA encoding an immunodominant myelin basic protein peptide targeted to Fc of immunoglobulin G suppresses experimental autoimmune encephalomyelitis. *J. Exp. Med.* **187:** 1543–1548
60. Weissert R., Lobell A., de Graaf K.L., Eltayeb S.Y., Andersson R., Olsson T., Wigzell H. 2000. Protective DNA vaccination against organ-specific autoimmunity is highly specific and discriminates between single amino acid substitutions in the peptide autoantigen. *Proc. Natl. Acad. Sci. USA.* **97:** 1689–1694
61. Tsunoda I., Tolley N.D., Theil D.J., Whitton J.L., Kobayashi H., Fujinami R.S. 1999. Exacerbation of viral and autoimmune animal models for multiple sclerosis by bacterial DNA. *Brain Pathol.* **9:** 481–493
62. Segal B.M., Chang J.T., Shevach E.M. 2000. CpG oligonucleotides are potent adjuvants for the activation of autoreactive encephalitogenic T cells *in vivo*. *J. Immunol.* **164:** 5683–5688
63. Davoust N., Nataf S., Reiman R., Holers M.V., Campbell I.L., Barnum S.R. 1999. Central nervous system-targeted expression of the complement inhibitor sCrry prevents experimental allergic encephalomyelitis. *J. Immunol.* **163:** 6551–6556