Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Potential Role of Paramyxoviruses in Multiple Sclerosis

Stuart D. Cook, M.D., Benjamin Blumberg, Ph.D.,† and Peter C. Dowling, M.D.‡*

Multiple sclerosis (MS) is the most common demyelinating disease of the central nervous system (CNS), with an estimated 500,000 individuals in the United States afflicted by this often debilitating disease. The etiology and pathogenesis of MS are unknown, although it is widely believed that one or more infectious agents trigger the disease, probably through an autoimmune mechanism.[14] Evidence for an infectious etiology of MS is indirect, as independent investigators have been unable to confirm any study claiming to have isolated or transmitted an infectious agent from MS tissue or identified foreign antigens in the CNS of MS patients, but not controls. However, if MS is an autoimmune disease initiated by a virus that is no longer present in the brain when neurologic symptoms first occur, studies searching for virus, viral antigen, or viral genome would be unrevealing.[14] At present, several lines of investigation—epidemiologic, serologic, and viral-induced animal models of demyelination—suggest that MS may be caused by a viral agent. In this article we shall review the evidence that two closely related morbilliform viruses, canine distemper virus (CDV) or measles virus (MV), could cause MS.

EPIDEMIOLOGIC STUDIES SUGGESTING AN INFECTIOUS CAUSE OF MS

The most convincing evidence that MS may be caused by one or more infectious agents comes from epidemiologic studies. These show a unique

*Professor and Chairman, Department of Neurosciences, University of Dentistry and Medicine, New Jersey Medical School, Newark; Chief, Neurology Service, Veterans Administration Medical Center, East Orange, New Jersey

†Associate Professor, Department of Neurosciences, University of Dentistry and Medicine, New Jersey Medical School, Newark; Chief, Neurovirology, Veterans Administration Medical Center, East Orange, New Jersey

‡Professor, Department of Neurosciences, University of Dentistry and Medicine, New Jersey Medical School, Newark; Assistant Chief, Neurology Service, Veterans Administration Medical Center, East Orange, New Jersey

worldwide geographic pattern of MS with zones of high risk roughly related to increasing latitude.[48] However, even within a given latitude, considerable variation in MS incidence can be seen. Migration of individuals from high- to low-risk areas indicates that exposure to an environmental factor, particularly during late childhood and early adult life, may be critical to the subsequent development of MS.[4] Further, recent epidemiologic data from the Faroe Islands[31, 50] and elsewhere[13, 15, 17, 21, 24, 49, 65] suggest that the time from exposure to the putative inciting factor and onset of neurologic symptoms may, in some instances, be shorter than previously thought: 5 years or less. It has also become apparent that MS is not always a stable endemic disease in a given locale; time clusters of MS have occurred in several isolated islands, and paradoxical increases or decreases in MS incidence may be seen in geographic areas of close proximity. For example, until recently the highest rate of MS in the world was noted in the Orkney and Shetland Islands off the northeast coast of Scotland, and it was thought that MS incidence had remained remarkably constant there for nearly a century.[63] In contrast, in the Faroe Islands, which are approximately 200 miles from the Shetlands, MS was not known to exist before 1943, and only one case of MS occurred there from 1961 to 1979.[50] However, in the interim period from 1943 to 1960, Kurtzke and Hyllested documented that 24 patients with MS had the onset of their neurologic illness in the Faroes.[50] Another North Atlantic island community, Iceland, has had two time clusters of MS: one beginning in 1921, the other during World War II.[17, 21, 49] More recently, it has been shown that MS incidence has been declining in the Orkney[22] and Shetland Islands[23] but increasing in the adjacent areas of Northeast Scotland[29] and Western Norway.[51] In considering what environmental factors might be responsible for the paradoxical MS experiences in Orkney, Shetland, and the Faroes, it is important to note that the climate in these islands and the genetic and socioeconomic backgrounds of the inhabitants are quite similar.[14] Variables such as hygiene, diet, temperature, or sunlight cannot be of primary importance in explaining the different MS experiences in Orkney, Shetland, and the Faroes.[14] The Faroe epidemic also indicates that genetic factors alone cannot explain the present low incidence rate of MS there because during the 18 years of the Faroe epidemic, the average annual incidence of MS was similar to other high-risk areas. One plausible explanation for the Orkney-Shetland-Faroes disparity in incidence of MS is that an infectious agent previously common in the Orkney and Shetlands was absent from the Faroes before World War II and since 1960, but was present there before or during the MS epidemic.[14] Any candidate agent suggested to be the cause of MS should lead to an explanation of the unique MS experiences in various parts of the world including the Orkney, Shetland, and Faroe Islands.

Although it is commonly thought that MS, like the Guillain-Barré syndrome (GBS), is an autoimmune demyelinating disease[16] caused by multiple infectious agents,[27] the virtual absence of MS before and after the 1943–1960 MS epidemic in the Faroes is perhaps more indicative of a single agent causing MS in this island. The unusual global distribution of MS and susceptibility primarily in young adults are also consistent with the possibility of a single agent as a primary cause of MS worldwide because

in GBS, known to be triggered by multiple infectious and noninfectious agents, a remarkably uniform baseline worldwide incidence is seen, as well as a wide age range of host susceptibility.[70] In summary, epidemiologic studies indicate that environmental factors, probably infectious, cause MS, and suggest that one agent may prove to play a major role.

EPIDEMIOLOGIC STUDIES LINKING CDV OR MV TO MS

It has been proposed that MS may, in some instances, be caused by canine distempter virus (CDV), either directly or through an autoimmune mechanism.[14] According to this hypothesis, exposure of susceptible individuals—possibly those with inadequate measles immunity—to CDV would result in MS occurring at variable times thereafter. The CDV-MS hypothesis, although controversial, has successfully predicted or can explain the unique MS experiences in the Faroes,[19] Iceland,[17, 21, 49] Orkneys,[22] Sitka, Alaska,[15] and the Avalon Peninsula of Newfoundland.[65] The outbreak of MS in the Faroe Islands from 1943 to 1960[50] was apparently preceded by a severe World War II epizootic of CDV in the native dog population. Neither CDV nor MS had been known to occur in the Faroes from 1920 to World War II, the former because a ban on importing dogs (which was not enforced during World War II) prevented CDV from entering these islands.[19] Distemper has not recurred in the Faroes since 1950, and only one case of MS was noted there from 1961 to 1977. The CDV-MS hypothesis has also retrospectively predicted time clusters of MS after severe, widespread CDV epidemics in Iceland in 1921 and 1941.[17, 21, 49, 54] In testing the CDV-MS hypothesis, a statistically significant higher annual incidence of MS per 100,000 population was found in Iceland in the 10 years after the 1921 CDV epidemic than in the 10 years before the epidemic. Similarly, significantly more MS was found after the 1941 CDV Icelandic epidemic than in the comparable time periods before the epidemic. Furthermore, changes in age-specific prevalence rates and age at onset of MS in Iceland from 1946 to 1965 were consistent with the possibility that many MS patients in this period had exposure to a World War II point infection that caused their neurologic disease.[17, 21, 49] Clustering of MS incidence was also predicted in Sitka, Alaska, shortly after a 1965 CDV epidemic there.[15] MS had not occurred at any other time between 1949 and 1979. More recently, the hypothesis predicted that MS would decline in incidence in the Orkney Islands, based on an apparent decline in CDV in the dog population after a 1959 CDV epidemic.[22] Since 1965, MS incidence rates have fallen significantly in Orkney compared with the incidence from 1941 to 1964. Alterations in age-specific prevalence, mean duration of illness, and mean age of the MS population are consistent with the decline in MS incidence in the Orkneys in recent years. In Newfoundland, independent retrospective observations on CDV animal incidence by veterinarians and MS annual incidence by neurologists reveal that on three occasions since 1968 there have been significant temporal associations between CDV outbreaks and subsequent local increases in MS incidence on the Avalon Peninsula.[65] The lag period between the outbreak of CDV and the subsequent increase in

local MS incidence was approximately 3 years on each occasion. Thus, there appears so far to be a close epidemiologic association between CDV infection in dogs and MS in man; epidemics of the former predicting increases in the latter where close contact between man and dog exists and vice versa.

No other factor has been put forth that can adequately explain even one of the MS experiences cited here. In particular, neither measles infection nor measles immunization alone are compatible with the unusual Faroe or Orkney MS experiences.[22, 50] Other epidemiologic evidence against measles as the sole cause of MS are the anecdotal reports of measles infection occurring for the first time after the onset of MS.[68] In at least one such instance, serologic evidence of an acute primary infection accompanied the typical clinical picture of measles. On the other hand, several surveys of childhood diseases indicate that patients with MS as a group have had measles at a later age than expected.[2, 3, 40, 74] In attempting to reconcile these seemingly contradictory epidemiologic data regarding measles and MS, one could speculate that if MS is caused primarily by one virus, then MV alone is not the cause, but decreased or altered measles immunity that in some instances may occur as a consequence of atypical age at MV infection may allow CDV, which is normally innocuous to humans, to trigger MS.

EPIDEMIOLOGIC CRITICISM OF THE CDV-MS HYPOTHESIS

The CDV-MS hypothesis has been controversial primarily because studies of previous exposure to dogs in patients with MS and controls have given conflicting results, with most studies failing to show more exposure to dogs in patients with MS (although many show a trend toward more exposure to dogs in the MS group).[10, 11, 42, 56, 62, 66, 75, 82] However, the background of dog ownership in controls in most studies has been high, ranging from 60 to 90 per cent. Because the risk of developing MS is probably quite low even when an individual is exposed to the factor(s) causing MS, the high level of exposure to dogs in the population makes retrospective small case control studies relatively insensitive for testing the CDV-MS hypothesis. In addition, the variable and often long latency before clinical manifestations occur in patients with MS makes it difficult, in retrospective case-control studies, to focus on a specific time before disease onset when exposure to dogs might have affected MS risk. On the other hand, even though distemper may be subclinical in up to 75 per cent of infected dogs,[6] several studies have shown increased exposure of MS patients to dogs with a distemper-like illness before onset of MS,[5, 24, 82] and if the CDV-MS hypothesis is correct, this is the more important relationship. Another criticism of the hypothesis is that MS occurs preferentially in temperate latitudes, whereas distemper, like measles, occurs ubiquitously. Although CDV occurs in all climates, the virus is heat sensitive, being more rapidly inactivated at warm temperatures.[6] Furthermore, clinical distemper peaks in cold, damp weather,[6] conditions under which dogs and people are more apt to be indoors in close contact.[5, 22, 56]

ANTIVIRAL ANTIBODIES IN MS SERUM AND CEREBROSPINAL FLUID

Serologic detection is based on the principle that most viruses elicit antibodies directed to antigenic components of the virus. Generally, the finding of high antibody titer in a single serum sample does not distinguish between recent and remote infection by that virus. However, if the viral-specific antibody of the IgM class is present, presumptive evidence of a recent or persistent infection by the agent can be inferred. In the absence of IgM antibody, it is necessary to demonstrate a 4-fold or greater change in antibody titer (rising or falling) in paired sera in order to have acceptable serologic evidence of active viral infection. In a chronic disorder such as MS, where exposure to the putative agent may have occurred many years earlier, even before neurologic symptoms were manifest, one would not expect to find diagnostic changes in serum viral antibody titer or increased viral specific IgM antibody.

Serology can be useful, however, in the study of large numbers of patients with a chronic disorder such as MS and an equal number of appropriately matched controls (age, sex, geographic area). Significantly higher antibody titer to a common virus in MS sera compared with controls would suggest the possibility of persistent infection by that virus in patients with MS. Even low levels of an antibody to an uncommon virus in MS but not control sera would implicate that agent as a possible cause of MS. However, in serologic surveys, one must be cautious in interpreting the meaning of titer differences between MS patients and controls, because HLA type,[80] high serum IgG, or an altered immune state may be associated with elevated viral antibody titers unrelated to the cause of disease. Failure to find higher antibody titers in MS sera than in controls would not exclude the possibility that a common virus causes MS (with MS being due to an unusual viral-host interaction). This is particularly true if the virus is no longer present when neurologic symptoms become evident and if CNS lesions are due to autoimmunity. Even a lower antibody titer in MS patients than controls could conceivably be found associated with a virus causing MS. For example, lower antibody titers to CDV are seen in animals with chronic CNS disease than in animals recovering from infection with this virus.[47] Lastly, if the etiologic agent is a weak antigen, or is closely related to another virus that is a better antigen, antibody might be present in low titer and only transiently, perhaps not being found several years later when neurologic disease begins. Thus, as Koprowski has pointed out, during a persistent virus infection, the host's antibody response may be bypassed, diminished, or restricted, and therefore the absence of an antibody response would not eliminate any virus(es) from being considered as possible etiologic agents in MS.[45]

MV Antibodies

Numerous surveys of antibody titers to viruses in MS have been carried out and reviewed extensively.[14] These studies have been directed primarily toward MV since the initial report of elevated serum antibody to MV in patients with MS by Adams and Imagawa in 1962.[1] Compilations of

these studies as of 1976 revealed serum antimeasles antibodies reported to be significantly increased in 31 of 35 publications.[57] This was true despite the use of at least eight different antibody detection systems and of antigenic preparations from at least three components of MV.[14, 57] It is unlikely, therefore, that the measles antibody detected represents cross-reacting antibody to nonmorbilliform antigens. The measles antibody titer elevation is modest, averaging only a one-tube (2-fold) increase over controls, and no relationship has been found between measles titer and disease activity or duration.[14, 20]

More recently, it has been shown that serum measles antibody in MS patients is probably directed to all the polypeptides of MV and thus does not resemble the serologic profile in subacute sclerosing panencephalitis (SSPE), in which decreased antibody to the M protein of MV is found.[38] Although increased serum antibody to the P protein of MV was found by Wechsler and Meissner in 5 of 24 MS patients but not controls, these results have not been confirmed by others.[84]

The measles serum antibody titer in siblings without evidence of MS also tends to be higher than that of controls.[14] Although this may reflect a common genetic pool that enables higher viral antibody titers to develop, it is also consistent with a shared environmental experience in affected and nonaffected siblings such as similar exposure to morbilliform viruses. High measles titers in non-MS siblings would also suggest that the finding of measles antibody elevation in MS patients is not totally an epiphenomenon to the MS process per se. The possibility that the increased measles antibody titer might reflect an environmental exposure is supported by the study of Panelius and associates,[59] who studied viral antibody titers in MS patients and controls in high-, low-, and intermediate-risk areas for MS. The greatest risk of developing MS was found where MV titers were highest in both MS patients and controls. Thus, siblings of patients with MS and unrelated controls in high-risk areas also have tendencies to high MV antibody titers.

Serum antibodies in MS patients have also been shown to be occasionally increased to vaccinia, rubella, herpes simplex, varicella zoster, Epstein-Barr virus, and other viruses, although far less consistently than to MV.[14]

Although there are fewer studies of viral antibody in cerebrospinal fluid (CSF) than in serum, similar results have been obtained.[36] Increased MV antibody in MS CSF has been found by investigators using different techniques and antigenic preparations.[14, 36] Furthermore, alterations in serum-CSF measles antibody ratios have been found that suggest that measles antibody is being produced in the central nervous system (CNS) or CSF.

Attempts have been made to relate oligoclonal bands in the CSF of patients with MS to measles antibody, analogous with the relationship between oligoclonal bands and MV in SSPE. However, it has not been possible to correlate MV-specific antibodies to the oligoclonal bands of IgG.[78] The measles antibodies are carried by different populations of IgG with differing migratory patterns on electrophoresis, and only a small portion of oligoclonal IgG can be removed from MS CSF after absorption with MV antigen preparations.[79]

As with serum studies, increased titers of CSF antibody and alterations in serum-CSF antibody ratios to other viruses have been described.[14, 36, 57] Although less frequent than measles, increased CSF antibody or abnormal serum-CSF ratios to vaccinia, herpes simplex, rubella, mumps, and other viruses have been reported in patients with MS.[14, 36] Furthermore, multiple virus antibodies may be seen in CSF from the same patient, and fluctuations in titer may occur. This raises the possibility that the alteration in viral antibody serum-CSF ratios may reflect in part an alteration in the blood-CSF barrier or may show that clones of cells continually producing antibody to multiple viruses are present intracranially.[14, 57]

CDV Antibodies

Relatively fewer serologic studies have been carried out searching for CDV antibodies in MS sera or CSF than for measles.[7, 8, 20, 33, 40, 42, 46, 53, 72] CDV serum neutralizing antibodies are elevated in MS patients as compared with controls in some, but not all, serologic surveys. However, the meaning of elevated CDV neutralizing antibody titer in MS patients is unclear, because MV neutralizing antibodies also tend to be elevated in the same MS sera, and there is a significant correlation between the antibody titers to the two viruses in both MS patients and controls.[8, 20, 53] This finding is consistent with the close antigenic (and genomic) relationship between CDV and MV.[9, 39, 71] One possible explanation for these serologic findings is that CDV neutralization is due solely to cross-reacting measles antibody and that CDV infection does not occur in humans (or MS patients). Another possible explanation for the elevated MV (and CDV) neutralizing antibody in MS sera is that CDV infection in humans occurring after measles infection or immunization causes a persistent amnestic reaction to MV with only a slight or transiently elevated antibody response to CDV. Such a phenomenon occurring with sequential exposure to closely related strains of influenza virus has been described and has been called the theory of original antigenic sin.[12] Precedence for an amnestic reaction to MV after CDV vaccination has been described in animals,[58, 67] including primates,[26] and the CDV antibody may, under certain circumstances, be of low titer and only transiently elevated (Fig. 1). For example, in a long-term pilot study, we vaccinated a monkey with MV in Freund's adjuvant, followed several months later by CDV (Fig. 1). After MV vaccination, there was an approximately 8-fold increase in MV neutralizing antibody to 1:8000 titer,* but no increase in CDV antibody titer. Over the next few months, the MV titer fell to less than 1:2000. However, after CDV vaccination, there was a persistent amnestic reaction to MV but only a transient increase in CDV neutralizing antibody lasting less than 2 months. The CDV neutralizing antibody was probably not just cross-reacting MV antibody because the titers of MV and CDV antibody were transiently the same at approximately 1:2000 immediately after the CDV vaccine, and there had been no particular increase in CDV antibody at other times when MV antibody titers were elevated. Thus, 2 years after the MV and CDV vaccinations, a persistently high MV antibody titer was present, but little CDV antibody, despite the

*Performed blindly by Dr. Max Appel, Ithaca, New York

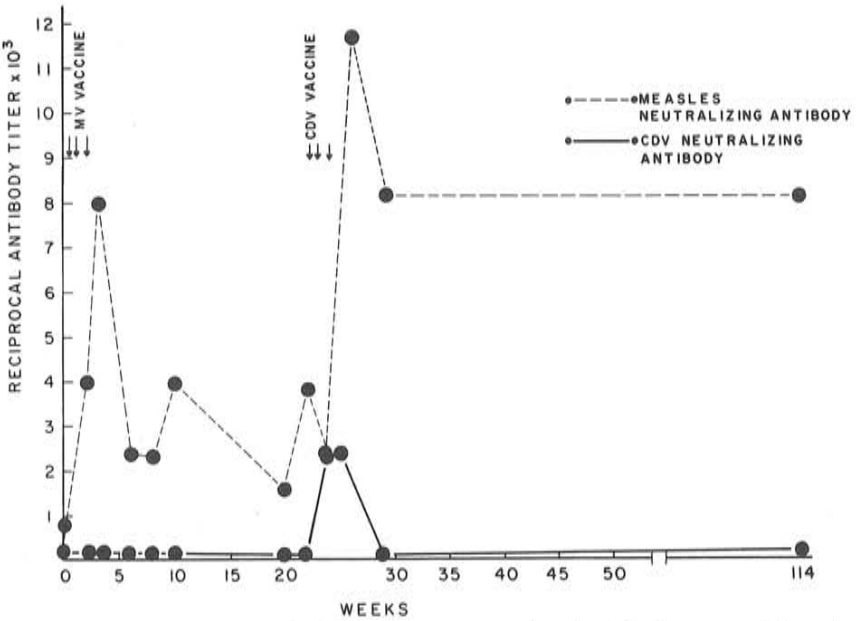

Figure 1. Neutralizing antibody response in primate (monkey 23) after sequential measles virus and canine distemper vaccinations: long-term study.

fact that this animal had previously reacted to both viruses. It should be noted that these serologic findings are not unlike those seen in MS patients, in whom the antecedent events are unknown, but elevated MV antibody is often present.

Serologic studies have also been carried out comparing MS serum and CSF with control fluids for the presence of immunoprecipitating antibodies to CDV and MV, using radiolabelled CDV and MV, and subsequent acrylamide gel electrophoresis of the precipitates. No difference between MS and control sera was found.[39, 46, 72] However, in one model study by Hall and Choppin, sera from animals immunized with MV precipitated all the polypeptides of CDV, indicating that it may be difficult (or impossible) with this technique to determine whether humans with prior measles infections have also been infected with CDV.[39] In another, unconfirmed study, using a similar technique, Krakowka and coworkers found significantly more MS than control CSF had anti-CDV P antibody, and a higher titer of anti-CDV P antibody was also found in the MS CSF, whereas no difference between MS and control CSF was noted for measles antibody to P.[46]

Attempts have been made to circumvent the problem of cross-reactivity by absorbing MS sera with MV and reassessing for CDV neutralizing antibodies.[7, 8] Although such tests showed marked reduction of the previously low-titered CDV neutralizing antibody, these assays cannot exclude the presence of low titer CDV antibody in MS sera, which cross-reacts with surface glycoproteins of MV and thereby would be removed by absorption with MV (or CDV). Indeed, CDV antibodies do react with most of the polypeptides of MV.[39, 71] Further, it has been shown that after

immunization with both MV and CDV, dogs develop serum antibodies with broader affinity for both viruses, so that after absorption of these sera to either MV or CDV, eluted antibodies still react with both viruses.[67] If, by analogy, man is exposed to both MV and CDV, it is conceivable that serum antibodies to MV and CDV could be absorbed by either virus, CDV antibody disappearing before MV antibody because of its lower initial titer. Still other attempts at distinguishing between MV and CDV antibodies have been carried out by exposing MS (and control) CSFs to various combinations of radioactively labeled and unlabeled CDV and MV, and subsequently determining affinity of the CSF antibodies for MV and CDV.[46] These assays have the same problem as the absorption studies in being unable to distinguish between cross-reactive antibodies with high affinity for both MV and CDV (which may be present in MS patients and normals) and CDV or measles-specific antibodies.

Thus, serologic tests to date have been unable to clearly answer the questions of whether CDV specific antibodies are present (perhaps in low titer) in any human sera, indicating that humans can be infected with CDV; and, in particular, whether CDV specific antibodies are present solely, or in higher titer in MS than control serum or CSF.

In summary, no single virus has been convincingly linked to MS by serologic testing at present. If MS is caused by one agent, then a morbilliform virus-like MV (or a closely related virus) is a leading candidate based on serology alone. In that case, the finding of elevated antibody to other viruses in MS sera and CSF could either be secondary to the MS process (perhaps associated with a defect in immunoregulation in MS) or due to a genetic difference between MS patients and controls, allowing higher viral antibody titers to be present.[80] A similar explanation could be used to explain why the oligoclonal bands in the CSF of MS patients are not primarily antibody to MV. Alternatively, one could propose multiple causes for MS to explain the varied serum and CSF findings or explain all the serologic abnormalities as being unrelated per se to the cause of MS.

VIRAL-INDUCED ANIMAL MODELS OF DEMYELINATION

Animal viruses such as CDV, the JHM strain of coronavirus, MV, Theiler's murine encephalomyelitis virus, Semliki Forest virus, and others can produce CNS demyelination in susceptible hosts.[14, 25] In many of these models, the mechanism of tissue damage is not understood but it appears that demyelination occurs in some instances by direct lytic effect of the virus and in others at least in part by immune mechanisms. The coronavirus JHM can cause a subacute demyelinating encephalomyelitis in rats.[83] Passive transfer of myelin basic protein–stimulated mononuclear cells from diseased animals (but not controls) to previously healthy recipients induced CNS histologic lesions and clinical signs in the recipients (but not demyelination). If confirmed, and the possibility of transfer of virus in mononuclear cells is excluded, this model would show that a CNS viral infection could potentially become a self-perpetuating autoimmune neurologic disorder and could have important implications for the pathogenesis of MS.

Both MV in man and CDV in the dog are naturally occurring demyelinating disorders. CDV is a ubiquitously distributed morbilliform virus, antigenically related to MV, but CDV is far more neurotropic in its natural host, the dog, than is MV in its natural host, humans. Some strains of CDV cause demyelination in 90 per cent of infected dogs,[41, 77] as compared with neurologic disease in approximately 0.1 per cent of humans with MV infection. The neurologic disorder in dogs may be acute or occur some months after an initially overt or subclinical system infection.[6] Early CDV-induced demyelination seems due to direct viral damage, but the role of the immune response in late demyelination is unclear.[25, 47] The neurologic disorder is quite variable and includes signs of optic neuritis, myelitis, ataxia, nystagmus, tremor, seizures, myoclonus, paresis, and psychic changes. Demyelinating lesions are similar to MS in many ways, the main exception being that virus can be found in the brains of infected dogs (albeit occasionally with some difficulty).[41, 77, 85] A persistent CNS infection may also occur and may be characterized by relapses and remissions, but long-term follow-up of these animals has been quite limited, and it is not known, for example, whether virus would persist in the CNS under these circumstances. Animals with chronic CDV infections of the CNS may have lower CDV antibody titer than those that fully recover.[47] The spectrum of host susceptibility to experimental nondemyelinating neurologic disease ranges from mice[52] to primates,[87] and a chronic remitting and exacerbating neurologic disorder in mice has been established after one intracerebral inoculation of CDV.[52] To date, there is relatively little evidence to support the possibility that CDV can infect man, although in one unconfirmed study, evidence for asymptomatic CDV infection in human volunteers was reported by Nicolle who was allegedly able to transmit CDV from human blood to dogs within 6 days of administering live virus to the volunteers.[55]

Although CDV is highly neurotropic, previous MV vaccination of dogs can decrease the neurologic complications of subsequent CDV infection.[6] Thus, there is a biologic precedence in which one of two closely related viruses protects against the neurotropic effect of the second. If CDV initiates MS, and the same immunologic relationship between measles and CD exists in humans as in the dog, high levels of measles immunity might prevent MS.

MV can induce experimental neurologic disease in animals ranging from hamster to monkey[60, 81] and occurs naturally in man.[43] CNS disease can occur through natural routes of spread of MV in both hamsters[60] and man,[43] and a demyelinating disorder, measles encephalitis, is seen in one of 1000 individuals infected with this virus.[43] Although there have been isolated reports of MV being cultured from the brain or CSF of patients with measles encephalitis,[76] it is commonly believed that measles encephalitis is an immunologically mediated disorder and that in most cases there is no evidence of viral invasion of the CNS.[43] Mild forms of measles encephalitis may be more common than is generally appreciated, as one third to one half of patients with uncomplicated measles have transiently abnormal electroencephalograms or pleocytosis in the CSF.[60]

SEARCH FOR MV AND CDV ANTIGEN, GENOME, AND VIRUS IN MS TISSUES

At one time or another, claims have been made to have isolated a large number of infectious agents, including MV, from MS brain specimens.[14] None of these claims has been confirmed by independent investigators, and in some instances, it appears that these isolated findings may have represented laboratory contaminants or spontaneous unrelated infections of laboratory animals, cell lines, or patients.

Studies using radioimmunoassay, immune electronmicroscopy, immunofluorescence, and immunoperoxidase techniques have been carried out in order to search for specific viral antigen in MS tissue.[14] These studies used high-titered viral antisera as probes. To date, no evidence of measles or CDV antigens have been found in MS brains.[18, 37] In considering the significance of these negative findings, it should be noted that such assays fail to detect less than 5 pfu of CDV per ml of tissue and were not sensitive enough to pick up MV in hamsters with chronic measles encephalitis.[18, 69]

In contrast to these results, Pertschuk and associates and Prasad and colleagues claimed to have identified measles virus antigen in 24 of 24 jejunal biopsies of MS patients and allegedly recovered paramyxovirus from six specimens.[61, 64] However, in their experiments, they used "inactivated" Sendai virus to fuse cells, and it is possible that the paramyxovirus isolated may actually have represented Sendai. More recently, Ebina and coworkers, using a polyethylene glycol cell-fusion technique, reported MV antigen recovery from jejunal mucosal biopsies of seven of 11 patients with MS after fusion with Vero cells, but not from six non-neurologic controls.[30] In these experiments, ultrastructural studies were not carried out, nor were cytopathic effects described. The demonstration by immunofluorescent technique of measles antigen in jejunal biopsies was not confirmed by Woyciechowska and associates in four measles antigen-positive biopsies supplied by Pertschuk.[86] Two other laboratories also failed to duplicate these results.[32, 44]

In 1981, Haase and colleagues reported finding MV nucleotide sequences in one of 4 MS brain specimens, using the sensitive technique of in situ hybridization.[35] Because CDV and MV genomes may cross-react under certain conditions in nucleic and hybridization studies,[9] one could not, on the basis of their report, exclude the possibility that Haase and coworkers were demonstrating CDV rather than MV genome. Subsequently, Haase and associates extended their studies and reported the presence of MV genomic sequence (or a closely related virus) in the brains of nearly half of patients with MS and also control brains.[34] This finding of MV genomic sequences in control as well as MS brain material could not be used as definitive evidence that MV caused MS, but if confirmed, would indicate that humans commonly harbor MV in their brains. However, studies by other laboratories, even using several different cloned C-DNA measles probes, have failed to confirm the findings of Haase and colleagues.[28, 73]

To date, there have been no published reports on attempts to find CDV genome in MS brain tissue by in situ techniques, although in our

laboratory, we have been unable to find evidence of CDV in MS brain using the technique of dot blot hybridization. In summary, there are no confirmed studies showing MV or CDV virus, viral antigen, or viral genome in MS tissues.

SUMMARY AND CONCLUSIONS

At the present time the cause of MS remains unknown, and it is unclear whether one or multiple agents trigger MS, although epidemiologic evidence seems to favor the former as being more likely. In considering the viability of measles or CDV as candidate agents in the causation of MS, it is worthwhile considering each agent alone with regards to the known MS serologic and epidemiologic data.

Measles Virus

If one agent causes most cases of MS, MV is the leading candidate agent at present on the basis of serology alone, although not all patients with MS have high MV antibody titers. Measles has been extremely common, although diminishing considerably since the widespread use of MV vaccine beginning about 1963. MV not infrequently causes an acute demyelinating disease in man, which may be immunologically mediated, and can also cause a chronic progressive neurologic disease (SSPE) in humans. As it is likely that MS is an unusual complication of a common viral infection, the fact that patients with MS sometimes have had MV infections at a later age than expected could be the unusual virus-host interaction responsible for initiating MS. The major barrier to the MV-MS hypothesis is epidemiologic. MV infection alone cannot explain the unique worldwide distribution of MS and is incompatible with both the observed incidence of MS in the Faroes from 1920 to 1979 and the decline in MS incidence in the Orkneys since 1965. Further, the recent increases in MS reported in Western Norway and Northeast Scotland have occurred despite a probable decline (or no increase) in MV infections in recent years, associated with more common use of MV vaccine. Thus we conclude that MV alone is unlikely to be the primary cause of MS, assuming one agent causes the majority of MS.

Canine Distemper Virus

In our opinion, CDV is the leading candidate agent for causing MS based on epidemiologic considerations alone. Although anecdotal, CDV infection is a plausible explanation for both the Faroe MS epidemic, and the virtual absence of MS before 1943 and since 1960 in these islands. Epidemics of CDV in the dog population successully predicted subsequent MS clusters in Iceland, Sitka, and Newfoundland. Further, a decline in CDV incidence in the Orkney Islands since 1959 successfully predicted a decline in MS there beginning in 1965. CDV in dogs is still quite common, although probably diminishing with the more widespread use of CDV vaccine. However, CDV vaccine is not as effective as many other vaccines, CDV infection occurring in previously vaccinated dogs, and major epidemics

of CDV still occur in areas with good veterinary care, for example, Switzerland, 1984 to 1985, and Australia, 1982 to 1984. An animal model of CDV exists that by natural routes of infection can produce a high rate of primary demyelination. Current serologic studies cannot be interpreted as either demonstrating or excluding the presence of CDV-specific antibodies in MS sera, and based on theoretical considerations, one could speculate that CDV is the stimulus for the increased MV antibody seen in patients with MS. The major objection to the CDV-MS hypothesis has been the lack of biologic evidence that CDV infects humans or causes MS. Thus, the evidence, to date, implicating CDV as the cause of MS is circumstantial. Nevertheless, based on the sum of evidence, we conclude that if MS is caused primarily by one agent, then CDV is the leading candidate agent in its causation at present. The CDV-MS hypothesis is testable in that it predicts that mandatory annual CDV vaccination of the dog population in a high incidence area of MS would lead to a decline in MS incidence within 10 years.

Possible MV-CDV Interaction

In considering the interrelationship between MV and CDV, it is apparent that these two viruses are extremely closely related antigenically and genomically, probably a reflection of a common viral ancestry. Previous MV immunity protects the dog against subsequent neurovirulence by CDV. If CDV can cause MS, and a similar immunologic relationship between these two closely related viruses exists in humans, as in the dog, then one could hypothesize that previous effective measles immunity might prevent MS from occurring in humans on exposure to CDV. Conversely, MS might occur in those individuals exposed to CDV who have impaired or altered measles immunity. It is plausible that an abnormally late age of MV infection could be one set of circumstances leading to such a state of altered measles immunity and be an important factor in determining the unfortunate minority who develop MS. If such an interaction between MV and CDV exists in humans, then increased measles immunity in man and decreased CDV in the dog population would lead to a decreased incidence of MS, whereas decreased measles immunity in man or increased CDV in canines would lead to an increase in MS incidence. Thus, widespread measles vaccination should lead to a decrease in MS, unless long-term measles immunity is diminished by this method of protection (as compared with natural infection) or unless this is counterbalanced by an increase in CDV infection in the canine population.

REFERENCES

1. Adams, J. M., and Imagawa, D. T.: Measles antibodies in multiple sclerosis. Proc. Soc. Exp. Biol. Med., 3:562–566, 1962.
2. Alter, M.: Is multiple sclerosis an age-dependent host response to measles? Lancet, *1*:456–457, 1976.

3. Alter, M., and Cendrowski, W.: Multiple sclerosis and childhood infections. Neurology, 26:201–204, 1976.
4. Alter, M., Leibowitz, U., and Speer, J.: Risk of multiple sclerosis related to age at immigration to Israel. Arch. Neurol. (Chicago), *15*:234–237, 1966.
5. Anderson, L. J., Kibler, R. F., Kaslow, R. A., et al.: Multiple sclerosis unrelated to dog exposure. Neurology (Cleveland), *34*:1149–1154, 1984.
6. Appel, M. J. G., and Gillespie, J. H.: Canine Distemper Virus. Vienna, Springer-Verlag, 1972, pages 1–153.
7. Appel, M. J., Glickman, L. I., Raine, C. S., et al.: Viruses and multiple sclerosis. Neurology, *31*:944–949, 1981.
8. Arnadottir, T.: Measles and canine distemper virus antibodies in patients with multiple sclerosis determined by radioimmunoassay. Acta Neurol. Scand., *62*:81–89, 1980.
9. Barrett, T., Underwood, B., and Mahy, B. W. J.: Morbillivirus genome structure and expression. Proceedings of International Conference in Virology, Catania, Sicily, 1985.
10. Bauer, H. J., and Wikstrom, J.: Multiple sclerosis and house pets. Lancet, 2:1029, 1977.
11. Bunnell, D. H., Visscher, B. R., and Detels, R.: Multiple sclerosis and house dogs: A case-control study. Neurology (NY), 29:1027–1029, 1979.
12. Coleman, M. T. and Dowdle, W. R.: Properties of the Hong Kong influenza virus: Part I. (General characteristics of the Hong Kong virus). Bull. W.H.O., *41*:415–418, 1969.
13. Cook, S. D. and Dowling, P. C.: A possible association between house pets and multiple sclerosis. Lancet, *1*:980–982, 1977.
14. Cook, S. D., and Dowling, P. C.: Multiple sclerosis and viruses: An overview. Neurology (NY), *30*:80–91, 1980.
15. Cook, S. D., and Dowling, P. C.: Distemper and multiple sclerosis in Sitka, Alaska. Ann. Neurol., *11*:192–194, 1981.
16. Cook, S. D., and Dowling, P. C.: The role of autoantibody and immune complexes in the pathogenesis of Guillain-Barré syndrome. Ann. Neurol., *9*(Suppl.):70–79, 1981.
17. Cook, S. D., Dowling, P. C., Norman, J., et al: Multiple sclerosis and canine distemper in Iceland. Lancet, *1*:380–381, 1979.
18. Cook, S. D., Dowling, P. C., Prineas, J. W., et al.: A radioimmunoassay search for measles and distemper antigens in subacute sclerosing panencephalitis and multiple sclerosis brain tissues. J. Neurol. Sci., *51*:447–456, 1981.
19. Cook, S. D., Dowling, P. C., and Russell, W. C.: Multiple sclerosis and canine distemper. Lancet, *1*:605–606, 1978.
20. Cook, S. D., Dowling, P. C. asnd Russell, W. C.: Neutralizing antibodies to canine distemper and measles virus in multiple sclerosis. J. Neurol. Sci., *41*:61–70, 1979.
21. Cook, S. D., Gudmundsson, G., Benedikz, J., et al.: Multiple sclerosis and distemper in Iceland: 1966–1978. Acta Neurol. Scand., *61*:244–251, 1980.
22. Cook, S. D., Cromarty, J. I., Tapp, W., et al.: Declining incidence of multiple sclerosis in the Orkney Islands. Neurology, 35:545–551, 1985.
23. Cook, S. D., McDonald, J., Tapp, W., et al.: A decline in incidence of multiple sclerosis in the Shetland Islands. Unpublished data, 1986.
24. Cook, S. D., Natelson, B. H., Levin, B. E., et al.: Further evidence of a possible association between house dogs and multiple sclerosis. Ann. Neurol., 3:141–143, 1978.
25. Dal Canto, M. C., and Rabinovitz, S. G.: Experimental models of virus-induced demyelination of the central nervous system. Ann. Neurol., *11*:109–127, 1982.
26. DeLay, P. D., Stone, S. S., Karzon, D. T., et al.: Clinical and immune response of alien hosts to inoculation with measles, rinderpest, and canine distemper virus. Am. J. Vet. Res., 26:1359–1373, 1965.
27. Dowling, P. C. and Cook, S. D.: Role of infection in Guillain-Barré syndrome: Laboratory confirmation of herpesviruses in 41 cases. Ann. Neurol., *9*(Suppl.):44–55, 1981.
28. Dowling, P. C., Blumberg, B. M., Kolakofsky, D., et al.: Measles virus nucleic acid sequences in human brain. Unpublished data, 1986.
29. Downie, A. W., and Pradse, J. G.: The chief scientist reports: Multiple sclerosis in North East Scotland. Health Bull., *42*:151–156, 1984.
30. Ebina, T., Tsukamoto, T. and Suzuki, H.: Measles virus in jejunum of patients with multiple sclerosis. Lancet, *1*:99, 1979.
31. Fischman, H. R.: Multiple sclerosis: A two-stage process? Am. J. Epidemiol., *114*:244–252, 1981.

32. Fraser, K. B., Haire, M., and Millar, J. H. D.: Measles antigen in jejunal mucosa in multiple sclerosis. Lancet, *1*:1313–1314, 1977.
33. Gorman, N. T., Habicht, J., and Lachmann, P. J.: Intracerebral synthesis of antibodies to measles and distemper viruses in patients with subacute sclerosing panencephalitis and multiple sclerosis. Clin. Exp. Immunol., *39*:44–52, 1980.
34. Haase, A. T., Stowring, L., Ventura, P., et al.: Detection by hybridization of viral infection of the human central nervous system. Ann. N.Y. Acad. Sci., *436*:103–108, 1984.
35. Haase, A. T., Ventura, P., Gibbs, C. J., Jr., et al.: Measles virus neucleotide sequences: Detection by hybridization in situ. Science, *212*:672–675, 1981.
36. Haire, M.: Significance of virus antibodies. Br. Med. Bull., *33*:40–44, 1977.
37. Hall, W. W., and Choppin, P. W.: Failure to detect measles virus proteins in brain tissue of patients with multiple sclerosis. Lancet, *1*:957, 1982.
38. Hall, W. W., Lamb, R. A., and Choppin, P. W.: Measles and SSPE virus proteins: Lack of antibodies to the M protein in patients with subacute sclerosing panencephalitis. Proc. Natl. Acad. Sci. USA, *76*:2047–2051, 1979.
39. Hall, W. W., Lamb, R. A., and Choppin, P. W.: The polypeptides of canine distemper virus: Synthesis in infected cells and relatedness to the polypeptides of other morbilliviruses. Virology, *100*:433–449, 1980.
40. Haile, R., Smith, P., Read, D., et al.: A study of measles virus and canine distemper virus antibodies and of childhood infections in multiple sclerosis patients and controls. J. Neurol. Sci., *56*:1–10, 1982.
41. Higgins, R. J., Krakowka, S. G., Metzler, A. E., et al.: Primary demyelination in experimental canine distemper virus-induced encephalomyelitis in gnotobiotic dogs: Sequential immunologic and morphologic findings. Acta Neuropathol. (Berl.), *58*:1–8, 1982.
42. Hughes, R. A., Russell, W. C., Froude, J. R., et al.: Pet ownership, distemper antibodies and multiple sclerosis. J. Neurol. Sci., *47*:429–432, 1980.
43. Johnson, R. T., Hirsch, R. L., Griffin, D. E., et al.: Clinical and immunological studies of measles encephalitis. Trans. Am. Neurol. Assoc., in press, 1986.
44. Kingston, D., Shiner, M., Lange, L. S., et al.: Measles antigen in jejunal mucosa in multiple sclerosis. Lancet, *1*:1313–1314, 1977.
45. Koprowski, H.: Search for viruses in multiple sclerosis. Neurology (Minneap.), *26*:80–82, 1976.
46. Krakowka, S., Miele, J. A., Mathes, L. E., et al.: Antibody responses to measles virus and canine distemper virus in multiple sclerosis. Ann. Neurol., *14*:533–538, 1983.
47. Krakowka, S., Olsen, R., Confer, A., et al.: Serologic response to canine distemper viral antigens in gnotobiotic dogs infected with canine virus. J. Infect. Dis., *132*:384–392, 1975.
48. Kurtzke, J. F.: Epidemiologic contributions to multiple sclerosis: An overview. Neurology, *30*:61–79, 1980.
49. Kurtzke, J. F., Gudmundsson, K. R., and Bergmann, S.: Multiple sclerosis in Iceland. I: Evidence of a post-war epidemic. Neurology (NY), *32*:143–150, 1981.
50. Kurtzke, J. F., and Hyllested, K.: Multiple sclerosis in the Faroe Islands. I: Clinical and epidemiological features. Ann. Neurol., *5*:6–21, 1979.
51. Larsen, J. P., Aarli, J. A., Nyland, H., et al.: Western Norway, a high-risk area for multiple sclerosis: A prevalence-incidence study in the county of Hordaland. Neurology (Cleveland), *34*:1202–1207, 1984.
52. Lyons, M., Hall, W., Cam, V., et al.: Induction of chronic neurologic disease in mice with canine distemper virus. Neurology, *30*:92–98, 1980.
53. Madden, D. L., Wallen, W. C., Houff, S. A., et al.: Measles and canine distemper antibody: Presence in sera from patients with multiple sclerosis and matched control subjects. Arch. Neurol., *38*:13–15, 1981.
54. Nathanson, N., Palsson, P. A., and Gudmundsson, G.: Multiple sclerosis and canine distemper in Iceland. Lancet, *2*:1127–1129, 1978.
55. Nicolle, C.: La maladie du jeune age des chiens est transmissible experimentalement a l'homme sous forme inapparente. Arch. Inst. Pasteur Tunis, *20*:321–323, 1931.
56. Norman, J., Cook, S. D., and Dowling, P. C.: Pilot survey of household pets among veterans with multiple sclerosis and age-matched controls. Arch. Neurol., *40*:213–214, 1983.

57. Norrby, E.: Viral antibodies in multiple sclerosis. Prog. Med. Virol., *24*:1–39, 1978.
58. Norrby, E., and Appel, M. J.: Humoral immunity to canine distemper after immunization of dogs with inactivated and live measles virus. Arch. Virol., *66*:169–177, 1980.
59. Panelius, M., Salmi, A., Halonen, P. E., et al.: Virus-antibodies in serum specimens from patients with multiple sclerosis, from siblings, and matched controls: A final report. Acta Neurol. Scand., *49*:85–107, 1973.
60. Parhad, I. M., Johnson, K. P., Wolinsky, J. S., et al.: Encephalitis after inhalation of measles virus: A pathogenetic study in hamsters. Ann. Neurol., *9*:21–27, 1981.
61. Pertschuk, L. P., Cook, A. W., and Gupta, J. K.: Measles antigen in multiple sclerosis: Identification in the jejunum by immunofluorescence. Life Sci., *19*:1603–1608, 1976.
62. Poskanzer, D. C., Prenney, L. B., and Sheridan, J. L.: House pets and multiple sclerosis. Lancet, *1*:1204, 1977.
63. Poskanzer, D. C., Prenney, L. B., Sheridan, J. L., et al.: Multiple sclerosis in the Orkney and Shetland Islands. I: Epidemiology, clinical factors, and methodology. J. Epidemiol. Community Health, *34*:229–239, 1980.
64. Prasad, I., Broome, J. D., Pertschuk, L. P., et al.: Recovery of paramyxovirus from the jejunum of patients with multiple sclerosis. Lancet, *1*:117–119, 1977.
65. Pryse-Phillips, W., and Jones, G.: Evidence for a temporal association between onset of multiple sclerosis and canine distemper in Newfoundland. J. Can. Neurol. Sci., *112*:329, 1984.
66. Read, D., Nassim, D., Smith, P., et al.: Multiple sclerosis and dog ownership: A case-control investigation. J. Neurol. Sci., *55*:359–367, 1982.
67. Roberts, J. A.: A study of the relationship between human measles virus and canine distemper virus. J. Immunol., *94*:622–627, 1965.
68. Ryberg, B.: Acute measles infection in a case of multiple sclerosis. Acta Neurol. Scand., 59:221–224, 1979.
69. Salmi, A., and Lund, G.: Immunoassay for measles virus nucleocapsid antigen: Effect of antigen-antibody complexes. J. Gen. Virol., *65*:1655–1663, 1984.
70. Schonberger, L. B., Hurwitz, E. S., Katona, P., et al.: Guillain-Barré syndrome: Its epidemiology and associations with influenza vaccine. Ann. Neurol., *9* (Suppl.):31–38, 1981.
71. Stephenson, J. R., and ter Meulen, V.: Antigenic relationship between measles and canine distemper viruses: comparison of immune response in animals and humans to individual virus-specific polypeptides. Proc. Nat. Acad. Sci. USA, *76*:6601–6605, 1979.
72. Stephenson, J. R., ter Meulen, V., and Kiessling, W.: Search for canine-distemper-virus antibodies in multiple sclerosis: A detailed virological evaluation. Lancet, 2:772–775, 1980.
73. Stevens, J. G., Bastone, V. B., Ellison, G. W., et al.: No measles virus genetic information detected in multiple sclerosis-derived brains. Ann. Neurol., *8*:625–627, 1980.
74. Sullivan, C. B., Visscher, B. R., and Detels, R.: Multiple sclerosis and age at exposure to childhood diseases and animals: Cases and their friends. Neurology (Cleveland), *34*:1144–1148, 1984.
75. Sylvester, D. L., and Poser, C. M.: The association of multiple sclerosis with domestic animals and house pets. Ann. Neurol., 5:207–208, 1979.
76. ter Meulen, V., Muller, D., Kackell, Y., et al.: Isolation of infectious measles virus in measles encephalitis. Lancet, 2:1172–1175, 1972.
77. Vandevelde, M., Higgins, H. J., Kristensen, B., et al.: Demyelination in experimental canine distemper virus infection: Immunological, pathologic, and immunohistological studies. Acta Neuropathol. (Berl.), *56*:285–293, 1982.
78. Vandvik, B., and Degre, M.: Measles virus antibodies in serum and cerebrospinal fluid in patients with multiple sclerosis and other neurological disorders with special reference to measles antibody synthesis within the central nervous system. J. Neurol. Sci., *24*:201–219, 1975.
79. Vandvik, B., Norrby, E., Nordal, H., et al.: Oligoclonal measles virus-specific IgG antibodies isolated by virus immunoabsorption of cerebrospinal fluids, brain extracts, and sera from patients with subacute sclerosing panencephalitis and multiple sclerosis. Scand. J. Immunol., 5:979–992, 1976.
80. Visscher, B. R., Sullivan, C. B., Detels, R., et al.: Measles antibody titers in multiple sclerosis patients and HLA-matched and unmatched siblings. Neurology (NY), *31*:1142–1145, 1981.

81. Waksman, B. H., Burnstein, T., and Adams, R. D.: Histologic study of the encephalomyelitis produced in hamsters by a neurotropic strain of measles. J. Neuropathol. Exp. Neurol., *21*:25–49, 1962.
82. Warren, S. A., Warren, K. G., Greenhill, S., et al.: How multiple sclerosis is related to animal illness, stress and diabetes. Can. Med. Assoc. J., *126*:377–383, 1982.
83. Watanabe, R., Wege, H. and ter Meulen, V.: Adoptive transfer of EAE-like lesions from rats with coronavirus induced demyelinating encephalomyelitis. Nature, *305*:150–153, 1983.
84. Wechsler, S. L., and Meissner, H. C.: Elevated antibody levels against measles virus P protein in sera of patients with multiple sclerosis. Infect. Immun., *40*:1226–1229, 1983.
85. Wisniewski, H., Raine, C. S. and Kay, W. J.: Observations in viral demyelinating encephalomyelitis canine distemper. Lab. Invest., *26*:589–598, 1972.
86. Woyciechowska, J. L., Madden, D. L., and Sever, J. L.: Absence of measles-virus antigen in jejunum of multiple-sclerosis patients. Lancet, *2*:1046–1049, 1977.
87. Yamanouchi, K., Yoshikowa, Y., Sato, T. A., et al.: Encephalomyelitis induced by canine distemper virus in non-human primates. Jpn. J. Med. Sci. Biol., *30*:241–257, 1977.

Department of Neurology
New Jersey Medical School
100 Bergen Street
Newark, New Jersey 07103