Multiple Sclerosis

Pathology, diagnosis and management

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Benign MS 148
Antibody was only found in the convalescent phase of the disease
Methylation of Myelin Basic Protein MBP 482
Corticospinal or pyramidal tract involvement 138
20% of autopsy proven MS not suspected 148
Cholesterol ester 364
Plaque grey and slightly pink in color 205 204
Edema 31
Freunds complete adjunct 480
Male Female ratio 74
Optic Neuritis 144 146 167
Twins 98
Seizures 142
Evoked potentials 196
Basic Protein removed from plaque 226 227
Lipid incorporation into myelin is slow
Macrophage 329 331 receptors 331
Light flashes 167
Unilateral visual loss so often seen with MS 152
Plasmapheresis 563
No antibodies 228
Damage from methyl group metabolism 498
Chiasma 164
MBP wow 133
Silver stain 205
Get article 411
Old name for Macrophages is Microglia 329
New Zealand 65
Reminiscent of lesions in methanol deterioration see article (407)
Foam cells 205
Vitreous Opacities 163
Demyelinating diseases 133
Platlet stikeyness
No antibodies 228
Lipid incorporation into myelin is slow
Humidity 520

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Multiple Sclerosis
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MULTIPLE SCLEROSIS /PLASMAPHERESIS /CHOLESTEROL-ESTER
MACROPHAGE /GREY-RED /PLAQUE
FOAM CELL /WOW /VITREOUS
SEIZURES

more recent plaques are less rubbery and may be rather grey 204
and slighyly pink in colour (see methanol plaque 4077 plaques
in the cortex and subpial regions of thegyri numerically outnumber
those in white matte by four to one. Periventricular plaques
(involving the lateral and fourth ventricles) are exceedingly 205
common and are found in over 90 % of cases of MS.Plaques are
also commonly found in the optic tracts and frequently in the
cord, pons, mid brain and basal ganglia. The frequent involvement
of the periventricular region calls for special comment: These
are usually old lesions part of the original pathogenic process.
The lesion wraps around one-third to one half of the ventricle;
the lateral and fourth ventricles are more commonly invloved
than the main body of the third ventricle. If the lesion is
still active some foam cells may be seen at its edge (pp205).
Conversion of cholesterol to cholesterol ester is considered
a secondary event and a function of enzymes in the mononuclear
phagocytes (pp225) Plasmapheresis caused subjective improvement
while 5-liter exchanges were performed every other day and
for one week afterward but improvement was not sustained when
exchanges began occuring every three to four weeks. Weekley
exchanges in conjunction with cyclophosphamid and prednisone
caoued improvement in all patients(pp563).
Digestion of myelin begins in Schwann cells and later in macrophages
that imvade the endoneurium (pp 30) .Negroes have only half
the risk as white males and young white females have twice
the risk of MS as do the white male.(pp74) classification of
demelinating diseases (pp133) . Corticospinal or pyramidal
tract involvement are commonly bilateral at an early stage-
other symptoms nystagmus and seizures pp138-145.vitreous opacities
-pp163. The macrophage is the most proment cellular representative
of the immune system in MS brain (pp330). hot and humid weather
can have worsening effect in some pp520. Most MS patients are
not habitual drinkers of abusers of alcohol (pp529).

macrophase — 336

indicate a relatively rapid movement throughout the sheath (Rawlins, 1973; Gould and Dawson, 1976). The idea that lateral diffusion of membrane components can occur provides a mechanism whereby some exchange between the postulated two metabolic compartments can take place. However, as electronmicroscope autoradiography studies suggest, not all of the components are equally free to diffuse and some lipid components, in particular, are more labile than the protein, a conclusion in line with the results of turnover studies of central myelin constituents (Benjamins and Smith, 1977).

It is worth pointing out that what is referred to as the 'fast' turnover rate in myelin still means a pool with a metabolic half life measured in days. This produces problems in interpreting isotope-labelling studies. For example, although the synthesis of lipids can be rapid (Jungalwala and Dawson, 1971), their incorporation into myelin is relatively slow (Benjamins and Smith, 1977). This means that myelin-labelling has to be examined in terms of the integral of the precursor specific activity with time, then some idea of the relative pool sizes can be obtained (Davison and Gregson, 1966). For myelin sulphatide the fast pool has a half life of 2–5 days, but accounts for only 0.2% of the total sulphatide. The small size of the dynamic pool would be appropriate to the morphological interpretation usually offered. Since the myelin-associated microsomal fractions, such as SN_4, are thought to represent the functional membrane regions, it might be expected that these fractions would metabolically reflect the dynamic 'fast' pools of myelin rather than the 'slow' pools. Certainly, it has been suggested that these 'myelin-like' fractions behave like myelin precursor material (Agrawal *et al.*, 1974).

In the adult animal, where myelin accretion can be considered to have ceased, then this metabolic activity is thought to represent an exchange process, presumably reflecting the 'instability' of the structure. Thus, as already stated, the lipid components appear to turnover faster than the protein components. However, this difference may of course be more apparent than real, in that the protein components may exchange and recycle without intervening degradation and resynthesis (Hennacy and Horrocks, 1978).

Interestingly, during Wallerian degeneration in the CNS the myelin is very slow to show either morphological (Cook and Wisniewski, 1973) or chemical change (Bignami and Eng, 1973), the time scale for change being in the terms of 50 to 100 days or longer (also see Adams, Chapter 8). If this behaviour reflects the 'intrinsic stability' of the myelin, then this would also accord with the calculated half lives for most of the constituents being in terms of 100–200 days (Benjamins and Smith, 1977). It appears that cellular activity is important for the rapid clearing of myelin debris (Hall and Gregson, 1977).

1.4.2 Synthesis and growth

During actual synthesis of the sheath, net accumulation of material must

occur. From isotope-labelling experiments it is apparent that a large proportion of the material synthesized enters the slowly metabolizing pool, a biphasic uptake not being apparent and the calculated half-lives being of the order of 100 days (Benjamins and Smith, 1977). Because there is growth, it is very difficult to investigate rapid turnover or exchange in the interparanodal myelin at this time.

It is now more widely accepted that the spiral form of the myelin sheath does not necessarily directly reflect movement of a cell or cell process during growth. It is suggested that the myelin membrane grows from the region of the external mesaxon by the transfer of materials synthesized in the perikaryon of the oligodendrocyte or Schwann cell (e.g. Rumsby, 1978). In general, lipids are understood to be synthesized in the smooth reticulum of the cell: microsomes from the brains of developing animals acquire a myelin-like lipid composition during the period of myelination (Bourre *et al.*, 1978). The immunochemical localization of myelin basic protein shows that this protein is concentrated in the oligodendrocyte during myelination, but is absent or below the limits of detection in the cell bodies of mature animals (Sternberger *et al.*, 1978; Hartman *et al.*, 1979); the same is true for proteolipid apoprotein (Agrawal *et al.*, 1977). The major myelin glycolipid galactosylceramide is present in the plasma membrane of the non-myelinating oligodendrocyte (Raff *et al.*, 1978) and in such membrane of the Schwann cell when under the influence of the axon (Mirsky *et al.*, 1980).

The 'myelin like' and SN_4 fractions have been regarded as myelin precursor membrane (Danks and Matthieu, 1979). The use of the word 'precursor' suggests that it is modified by the insertion of myelin proteins and lipids, and by the loss of other constituents. It then becomes the typical myelin membrane, which grows into the spiralled compacted myelin segment (see Rumsby, 1978). The growth of the sheath in this way requires that the membrane already spiralled around the axon must be able to accommodate the newly added material. There are no theoretical objections to this additional membrane being accommodated by a tangential slippage or shear at the surface, since the shearing of charged membrane surfaces involves very low forces (Gingell, 1971). This kind of process is quite feasible at the intraperiod line, but slippage may be more difficult at the period interface, where compaction appears to be more stable and protein is considered to act as a cross-linking agent (*vide infra*). It is possible, therefore, that the compact membrane expands in the form of a fused double membrane. However, in early myelin it is possible that shear can occur at both surfaces of the membrane, if the membrane is protein-deficient and, thus, not so strongly cross-linked. However, this kind of explanation does not fit all of the observations. Thus, although it can readily explain the increase in girth of the sheath, it does not explain how the deeper inner layers of membrane can become extended during longitudinal growth of the internode; the length of the paranodal region cannot alone account for the observed increment in

breakdown of myelin, a physical disruption of myelin lamellae and a chemical degradation beginning about one week after axonal degeneration (Majno and Karnovsky, 1958). Electron-microscopic observations indicate that the digestion of myelin begins in Schwann cells and later in macrophages that invade the endoneurium (Nathaniel and Pease, 1963). In the central nervous system, the breakdown of myelin occurs at about equal speed in peritraumatic regions or at the margin of infarcts, i.e. in the presence of macrophages that are derived from invading monocytes or microglial cells. At a distance from the area of injury the sheaths of degenerating tracts disintegrate more slowly (McCamen and Robins, 1959; Lampert and Cressman, 1966). Histochemically, myelin debris can be demonstrated for more than a year after transection of nerve fibres in the brain and spinal cord (Glees, 1943; Smith, 1951). The slower breakdown of central myelin is explained by the fact that sheaths in the central nervous system are not enclosed within glial cells. Phagocytosis and digestion of myelin require the presence of macrophages which abound next to necrotic tissue but are scanty in degenerating tracts. Collapsed loops of myelin are seen for weeks within gliosed, degenerated tracts without evidence of phagocytosis (Fig. 2.1a). Some of the sheaths show separation of myelin lamellae but most still reveal the normal periodicity of compact myelin. Well-

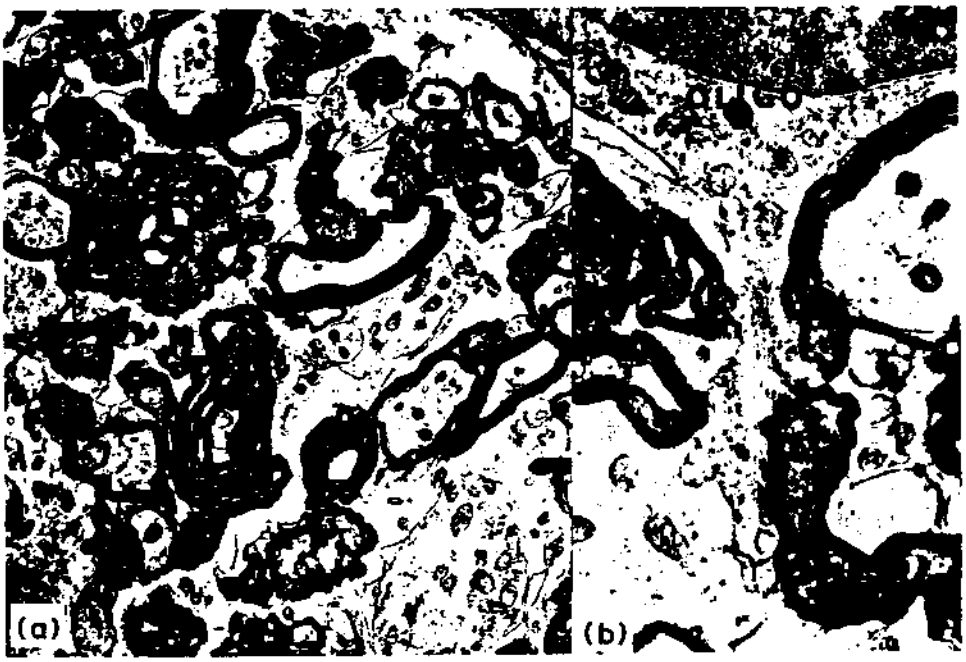

Fig. 2.1 a, b Axonal degeneration in thoracic cord of a rat 33 days after transection of the dorsal columns. a Myelin sheaths have collapsed around the degenerated axoplasm (×5100). b Oligodendrocyte adjacent to collapsed myelin sheaths show no phagocytic activity (×8500).

preserved oligodendroglial processes remain attached to collapsed myelin showing no evidence of phagocytosis (Fig. 2.1b). At later stages some myelin fragments are encountered within the cytoplasm of astrocytes. Occasional macrophages engulf compact myelin by penetrating between myelin lamellae or by entering spaces created by the disintegration of degenerated axoplasm. In this fashion macrophages are able to get into empty tubes of myelin, i.e. phagocytes are found completely surrounded by compact myelin. Such peculiarities were observed in degenerating tracts by Ramon y Cajal (1928) and later, confirmed by electron microscopy (Gray and Hamlyn, 1962; Lampert, Vogel and Zimmerman, 1968). Within macrophages myelin undergoes chemical degradation that is reflected by morphological changes as described below.

2.3 HYDRATION OF MYELIN SHEATHS

Two distinct alterations causing a swelling of myelin sheaths are recognized, a uniform and even separation of myelin lamellae and a very wide distension of sheaths due to the accumulation of fluid between lamellae.

A variety of disturbances have produced the uniform and even loosening of myelin lamellae (Fig. 2.2a). Exposure of sheaths to hypotonic solutions may cause this effect (Robertson, 1958). Water molecules attach to the lipoprotein layers of the intraperiod or minor dense lines splitting them apart. Similar lamellar separation has been described in oedema of cerebral white matter (Hirano, Zimmerman and Levine, 1965). Injury to myelin supporting Schwann cells have produced the same change in sheaths of peripheral nerves notably in metachromatic leukodystrophy (Lampert and Schochet, 1979) and in organotypic cultures after X-irradiation (Masurowsky, 1967). In the central nervous system swollen sheaths with uniformly separated lamellae have been observed in experimental allergic encephalomyelitis (EAE) (Lampert, 1967). The change affects most frequently the outer myelin lamellae. The separation begins with a split of the intraperiod line resulting in interlamellar spaces that are continuous with the extracellular space via the external mesaxon or between glial loops at nodes of Ranvier. The separation of the lamellae increases the width between major dense lines to about 25 nm or about twice that of compact myelin. The change may proceed to involve all lamellae and further result in splits of the major dense lines. The same alterations can be produced *in vitro* by exposure of myelin sheaths to heat-inactivated sera from animals with EAE suggesting that antimyelin antibodies may be responsible for this effect (Bornstein and Raine, 1976; Raine *et al.*, 1978). Immunoperoxidase studies have confirmed the presence of deposits of immunoglobulins in widened interlamellar spaces in similar experiments *in vitro* (Johnson, Raine and Bornstein, 1979). In demyelinating lesions in EAE one finds deposits of plasma precipitates which presumably contain immunoglobulins in close contact with sheaths that show this uniform swelling (Fig. 2.2b). In EAE the

included. Whether this holds true for the Portuguese rate as well is conjectural. In Iceland the rate was only 0.3 per 100 000. This low rate apparently contrasts with the high prevalence of MS in Iceland to be described below, even though the upper 95% confidence limit on this rate is about 0.9. Actually, MS in Iceland appears to have in fact changed in frequency, as will also be discussed later. We shall see that there is support for a rather low death rate in the 1950s.

Within Europe there seemed to be a sharp drop between the rates in the north and those for the Mediterranean basin. South American rates were rather low, as were those for US non-whites (of whom more than 90% are black or Negro). The Asian and African rates were clearly the lowest recorded. How accurate may be all these inferences as to the distribution of MS must await consideration of the morbidity data.

3.2.2 United States death rates

The American Public Health Association had sponsored a series of monographs based upon special tabulations of deaths in the US for 1959–61; one of these concerned neurological diseases, including MS (Kurland, Kurtzke and Goldberg, 1973). The average annual age adjusted death rate for MS was 0.8 per 100 000 population, with a slight female and a marked white

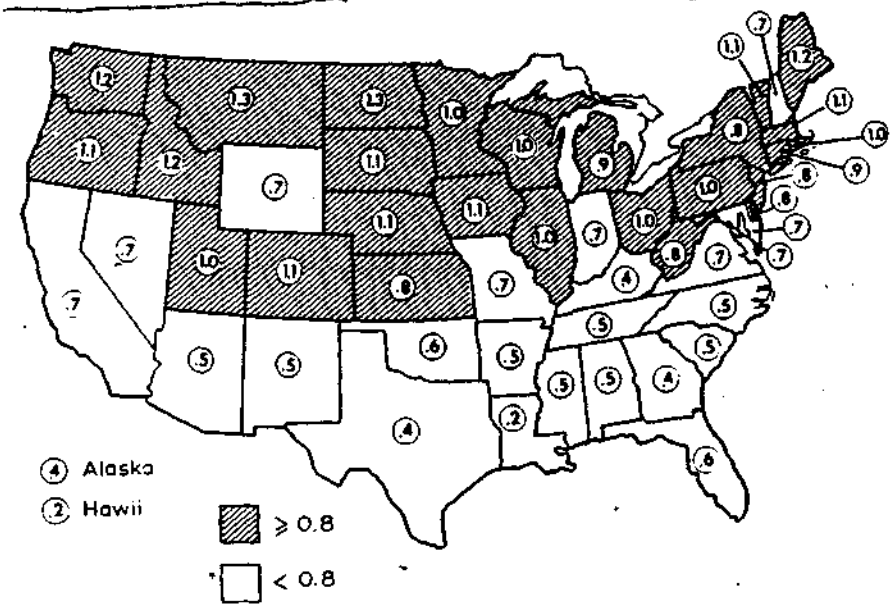

Fig. 3.4 Average annual age adjusted death rates for multiple sclerosis per 100 000 population by state of residence at death: United States, 1959–61. From Kurtzke *et al.* (1971).

preponderance. The male:female ratio on the rates was 0.9; the white:non-white ratio was 1.6. Both findings were consistent by geographical 'census' region of the US. Crude annual death rates were essentially stable over the 1949–1967 period. For 1959–61, among both males and females age adjusted death rates for MS were nearly three times higher for those who were single or divorced at time of death than rates among the married. Rates for the widowed were intermediate. Males had higher death rates than females among the widowed and divorced. Below age 65, female rates were higher than male for the married and were even more markedly in excess for the single. Geographically, all states south of the 37° parallel of north latitude showed low death rates (mostly 0.3–0.5), while almost all states to the north of this line were well in excess of the national mean (Fig. 3.4). This held true for residence at birth as well as at death, and for whites alone as well as for all residents. There was little consistent difference in MS death rates between urban and rural counties within the respective census regions, though for whites the urban rates tended to be somewhat higher.

3.3 GEOGRAPHIC DISTRIBUTION OF MS FROM PREVALENCE DATA

3.3.1 Europe

Prevalence studies provide our best information on the frequency of disease. However, they are expensive in time, people and money. Despite this, there are now well over 200 such surveys for MS. Almost all of them have been performed since World War II. Recently I tried to collect these studies and to rate them in terms of quality (Kurtzke, 1975; 1980c). It is obviously impractical here to list each of them. In the references cited are tables which define for each survey the author, the survey site, its latitude and longitude, the prevalence day, population, number of cases, the prevalence rate and its 95% confidence interval, and a rating as to the quality and hence comparability of the study. Class A studies had published data to indicate that they appeared reasonably complete as to case ascertainment, that they had followed appropriate survey methodology (Kurtzke, 1977), and that they used defined diagnostic criteria. Class B works were generally well done but had some features that might limit comparability—such as a survey performed because the area was thought exceedingly high or low in MS frequency, or lack of detail when this was one part of a broader group (e.g. some from Japan and Italy) that overall seemed acceptable. Class C surveys were clearly not comparable to the others, mostly because of major defects in case ascertainment. Class E works provided an *estimate* of MS prevalence from hospital and clinic case series. This estimate was obtained by taking the ratio of cases of MS to cases of amyotrophic lateral sclerosis (ALS) seen in the same interval, and then multiplying this ratio by a rate of 5 per 100 000, which was used as the 'standard' prevalence rate for ALS. The surveys were assigned

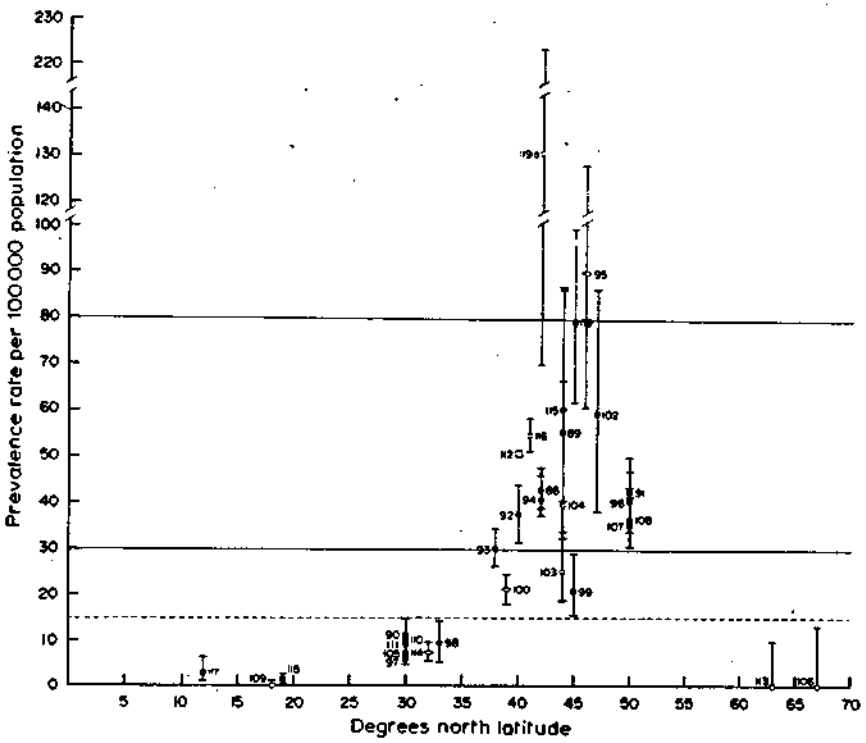

Fig. 3.10 Prevalence rates per 100 000 population for probable MS in the Americas, correlated with geographical latitude. Numbers identify the survey in Kurtzke (1975). Solid circles represent Class A studies, open circles Class B, diamonds Class C, and squares Class E. Vertical bars define 95% confidence intervals on the rates. From Kurtzke (1975).

Western Europe. Note that there were no studies from South America. More recent data confirm the North American rates, and there are now several MS/ALS ratio estimates for Argentina and Uruguay, and for Lima, Peru, which indicate these are medium frequency areas. Similar material for Venezuela and Brazil apparently allots these regions to the low frequency zone (Kurtzke, 1980c).

3.3.2 (a) US veteran series

The modest number of studies noted in Fig. 3.10 for the United States leaves much of the country undefined as to the distribution of MS. However, our recent history has provided us with a truly unique series. During World War II some 16.5 million Americans were in military service, and another 5 million served in the Korean Conflict (Kurtzke, 1978a). Legislation in this country has established multiple sclerosis as a 'service-connected' illness if manifestations of the disease were noted during military service or within

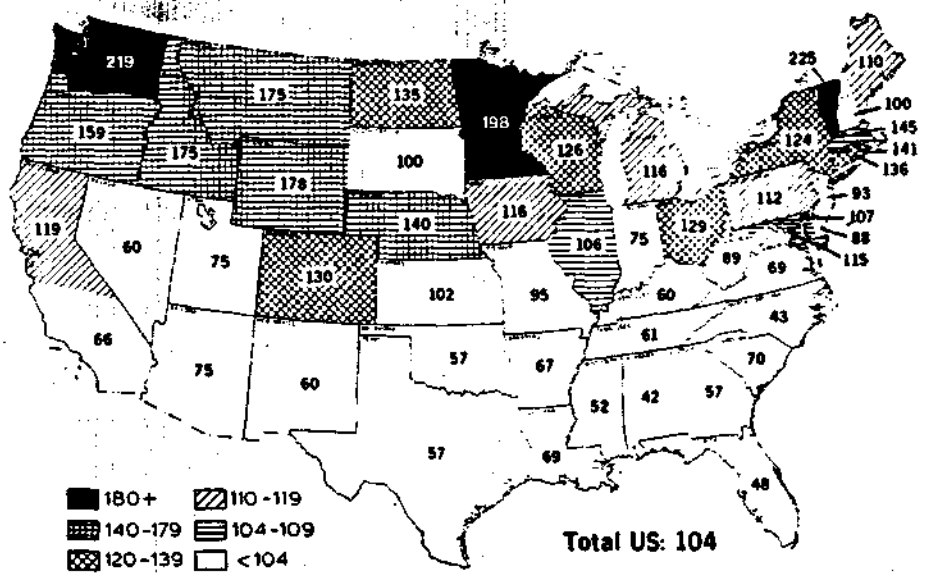

Fig. 3.11 Case-control ratios for US white male veterans of World War II service-connected for MS, according to state of residence at entry into military service. Ratios below 75% are medium frequency. Modified from Kurtzke (1978a).

seven years after discharge. We have identifed 5305 such veterans service-connected for MS. In our diagnostic review of a random sample of these cases, 96% met the clinical criteria of the Schumacher Committee (Schumacher *et al.*, 1965) for 'definite MS'. Each of the 5305 MS patients was matched to a *military* peer on the basis of age, date of entry and branch of service, and survival of the war. This provided us with an unbiased, pre-illness case control series of nationwide composition and unprecedented size (Kurtzke, Beebe and Norman, 1979a). Figure 3.11 shows the distribution of MS for white male veterans of World War II according to state of residence at entry into service, expressed as case control ratio percentages. From calculations described elsewhere (Bobowick *et al.*, 1978), the national case control ratio of 1.04 (or 104%) was estimated to be equivalent to a prevalence rate of 41.6 per 100 000. This in turn would define regions of less than 30 prevalence as those of less than 0.75 ratio. As may be seen in Figure 3.11, all states below the 37° parallel of north latitude would then fall within the medium frequency zone. Arizona's ratio was exactly 0.75 but was based on only 9 MS and 12 controls. Note how similar is this nationwide distribution to that from the US death rates in Fig. 3.4.

All states (and northern California) above the 37° parallel fall into the high frequency zone, except for Virginia (0.69 for 51 MS vs 74 controls) and Kentucky (0.60 for 37 vs 62). In the east, then, the high-to-medium dividing

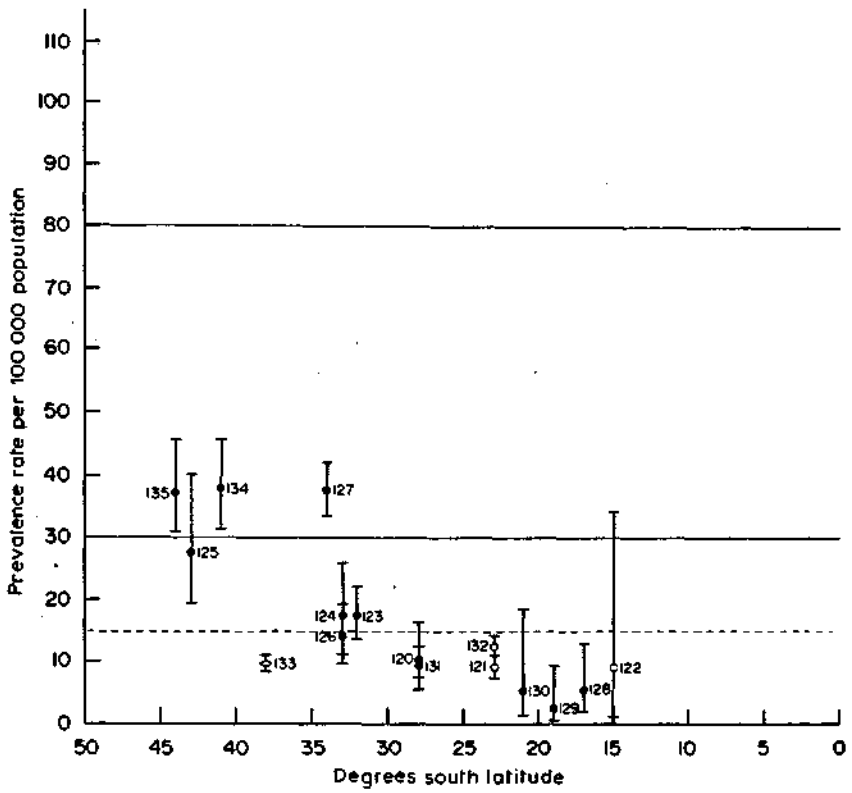

Fig. 3.12 Prevalence rates per 100 000 population for probable MS in Australia–New Zealand, as in Fig. 3.10. From Kurtzke (1975).

line passes the 39° parallel. The low ratio of 0.60 for Nevada in the west can be ignored since it came from only three MS and five controls. We therefore have in the United States too a quite sharp division between high and medium frequency bands.

3.3.3 Other regions

3.3.3 (a) Australia–New Zealand

To date we have no information on these countries additional to that presented in Fig. 3.12 (Kurtzke, 1975). At that time it seemed that Australia–New Zealand comprised principally a high frequency zone for 44°–34° south latitude, and a medium frequency region for 33°–15° south The recorded rates which were considered high were toward the lower end o this range. Geographically, this high zone included all of New Zealand as we as south-eastern Australia including Tasmania. The greater part of th continent was of medium frequency. New data should be available f

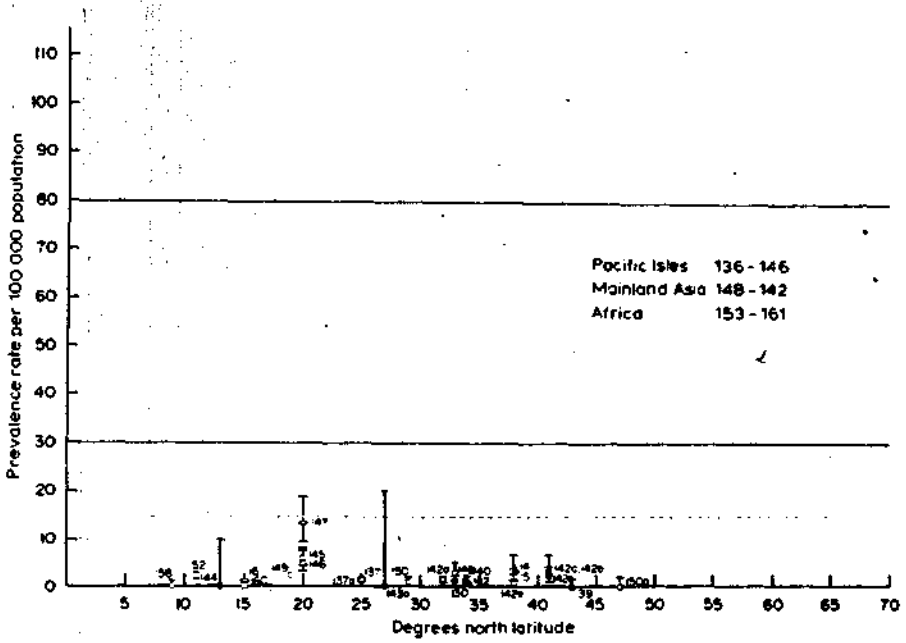

Fig. 3.13 Prevalence rates per 100 000 population for probable MS in Asia and Africa (northern hemisphere), as in Fig. 3.10. From Kurtzke (1975).

Australia in the near future, since a nationwide prevalence survey is now under way in that land.

3.3.3 (b) Asia and Africa

Rates from Asia and the Pacific in the northern hemisphere were all low, except that Hawaii (nos. 145, 146) may be in the medium zone (Fig. 3.13). These study sites extended from 8° to 47° north latitude. Later hospital series in Asia and additional prevalence studies in Japan indicate that there is no site in Asia thus far demonstrated to have more than a low frequency for MS (Kurtzke, 1980c).

In the southern hemisphere, with surveys from 30° to 6° south, all rates from Asia and Africa were also low, except for English-speaking native-born whites (no. 156) of South Africa (Fig. 3.14). Their rate of 11 contrasted with that of 3 for the Afrikaans-speaking native-born whites, a difference still without an explanation. It should be noted, though, that over the entirety of this vast continent there were data otherwise available only for Ethiopia, Natal and Senegal. In particular, there was no information on the Mediterranean littoral, where some recent hospital data from Tunisia *suggest* a prevalence rate within the medium risk range (Kurtzke, 1980c).

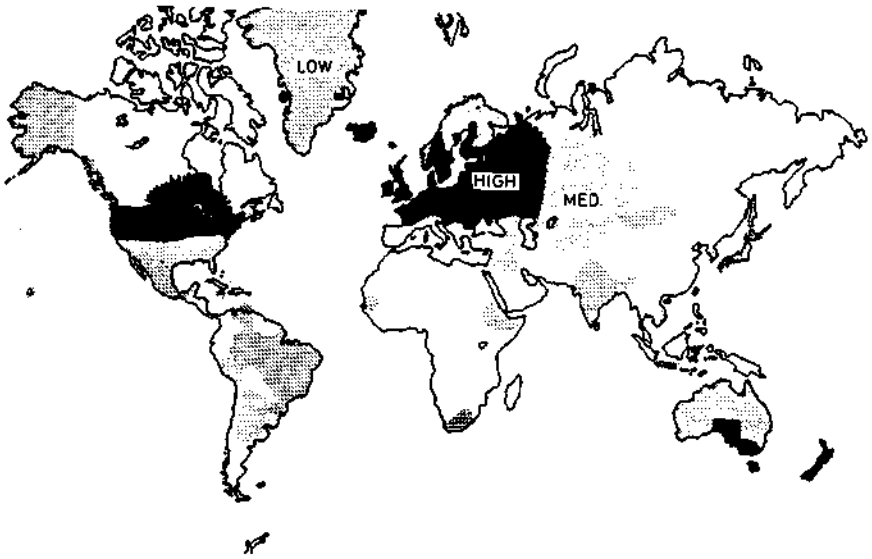

Fig. 3.15 World-wide distribution of MS as of 1980. High frequency areas are indicated in black, medium frequency areas with dots, and low frequency areas with diagonal dashes. Open areas are regions without data. South American frequencies are tentative. From Kurtzke (1980c).

frequency areas, with prevalence rates below 5 per 100 000, comprise all other known areas of Asia and Africa, Alaska and Greenland, and the Caribbean region to include Mexico and probably northern South America.

3.4 MS: AGE, SEX, RACE, AND FAMILY

3.4.1 Mortality data

Not only can we contrast death rates for geographical inferences, we can also see whether this terminal part of the illness provides us with useful information as to predilections by sex or race, as well as, of course, the age distributions themelves. Recall, though, that death rates as routinely published reflect the decision that this entity was the underlying cause of death, and special efforts are required to ascertain that proportion of deaths where the disease is listed as a contributory cause or associated condition on the death certificate. In Norway, Denmark and the United States, about three-quarters of deaths with MS listed were coded as underlying cause, but there were notable differences among the countries by age and sex as to these proportions, with Norway generally intermediate between the US and Denmark (Kurtzke, 1972a).

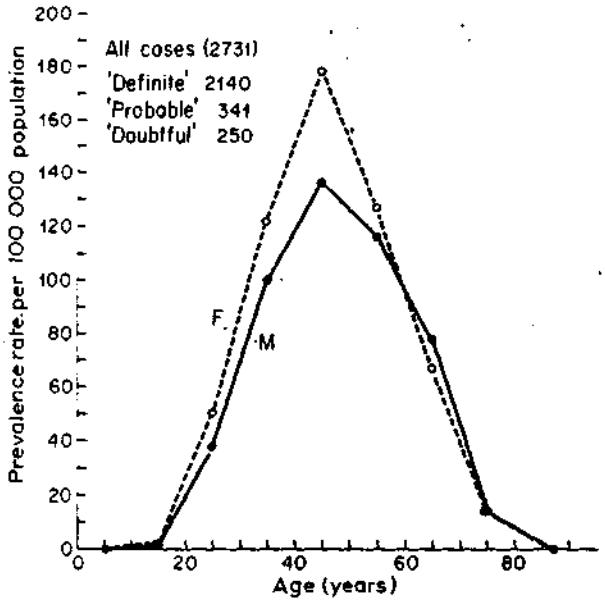

Fig. 3.17 Age and sex specific prevalence rates per 100 000 population for multiple sclerosis in Denmark, 1949, from data of Hyllested (1956). From Kurtzke and Kurland (1973).

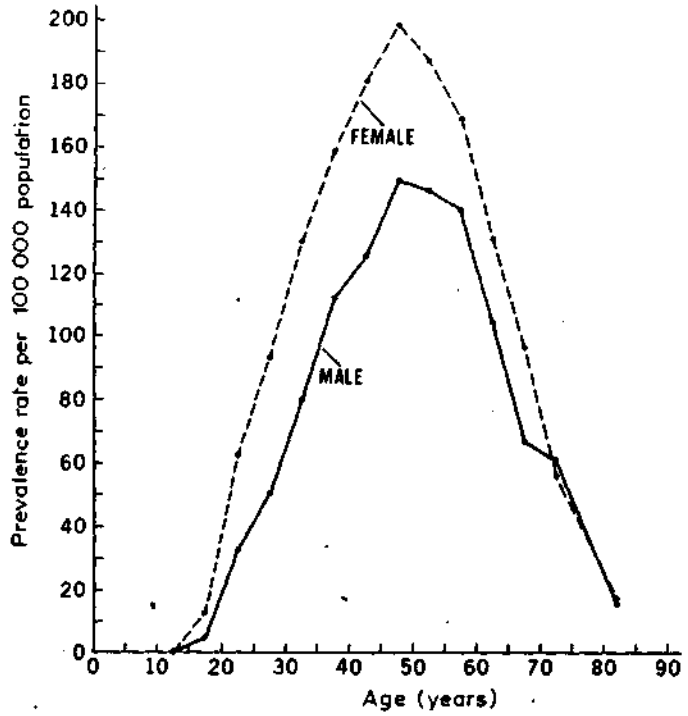

Fig. 3.18 Age and sex specific prevalence rates per 100 000 population for multiple sclerosis in Ireland, 1971, from data of Brady *et al.* (1977).

sex specific prevalence rates for probable MS in Ireland as of 1971, drawn from the data of Brady *et al.*, 1977). Note the similarity in all respects to Fig. 3.17. Either one provides a model for MS prevalence by age and sex in a high-frequency region.

3.4.2 (b) Incidence

By taking distributions at onset by age and sex for the Danish prevalent cases of definite and probable MS as of 1949, the population distribution of Denmark for 1940, and the average number of incident cases for 1939–45 (128.86 per year), it was possible to reconstruct age and sex specific annual incidence rates for MS (Kurtzke, 1969; Kurtzke and Hamtoft, 1976). These rates are drawn in Fig. 3.19, and demonstrate the female excess in the young and the maximal incidence at age 25–29. The annual incidence rate, all ages, was calculated as 3.35 per 100 000 population (3.00 male and 3.69 female). Here too the configurations do not suggest the need for age-adjustment in the usual material. For no source, mortality or morbidity, is there need to separate the data by sex, even though there is a modest female excess, particularly in the young.

We have seen above that Japan is a low-risk area, with a likely overall prevalence rate for probable MS of about 2 per 100 000 population. Shibasaki, Okihiro and Kuroiwa (1978) have pointed out the similarities in age at onset, course and duration of MS between orientals and whites in

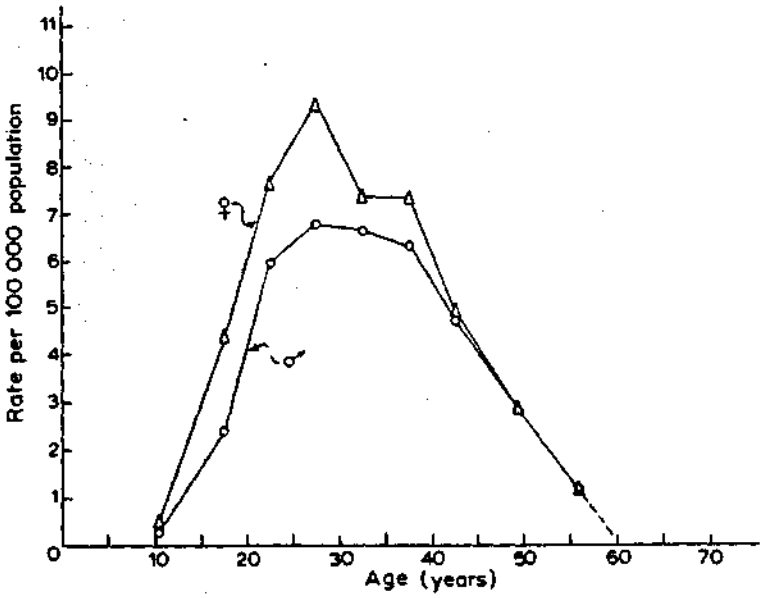

Fig 3.19 Average annual age and sex specific incidence rates per 100 000 population for multiple sclerosis in Denmark. From Kurtzke (1969).

Table 3.2 Multiple sclerosis: case/control ratios for 'other' males by birthplace and race, entire series. From Kurtzke *et al.* (1979a)

Birthplace and race	Ratio	Case/Control Total	N†	S†
Conterminous United States	0.48	11/23*	6/12	5/11
Amerindian	0.38	3/8	3/6	0/2
Mexican—Spanish American	0.60	6/10	1/1	5/9
Japanese	0.50	2/4	2/4	0/0
Mexico, Latin America, total	0.29	6/21		
Mexican—Spanish American	0.00	0/5		
Puerto Rican	0.38	6/16		
Hawaii, total	0.00	0/15		
Japanese	0.00	0/10		
Other	0.00	0/5		
Asia, total	0.00	0/14		
Chinese	0.00	0/4		
Filipino	0.00	0/9		
Other	0.00	0/1		
Total	0.23	17/73		

* Includes 1 Filipino control.

† N = Northern and middle tier of birth. S = Southern. For white males the MS/C ratios are 1.2 N and 0.6 S.

for MS have predominantly white populations. Regardless of residence in the US, in our veteran series blacks or Negroes have only half the risk of white males (Table 3.1). Note too that these young white females have nearly twice the risk of MS as do the white males. The group consisting of the 'Other' races suggests a paucity as well in American Indians and in Orientals (Table 3.2). Detels *et al.* (1977) in California have presented good evidence for a low prevalence among Japanese-Americans. The apparent deficit we found among Spanish-Americans would seem more a reflection of geography than race. This is borne out when comparisons by race are made among the foreign-born cases in the veteran series (Table 3.3). The deficit in the first two groups is equal for each race. Japanese and possibly Polynesians in Hawaii are low, as are Filipinos in the Philippines.

MS then is predominantly the white man's burden. However, it is clear that, where there are good data, the less-susceptible racial groups do share the geographical gradients of the whites, with higher frequencies in high-risk areas than in low. This observation differs from that of Alter and Harshe

Table 3.3 Multiple sclerosis: case/control ratios according to race and birthplace in selected regions, entire series. From Kurtzke *et al.* (1979a)

Region	Ratio	Case/Control			
		Total	White	Black	Other
Mexico, Central America	0.14	2/14	1/9	1/0	0/5
Puerto Rico	0.42	14/33	6/14	2/3	6/16
Hawaii	0.06	1/16	1/1	0/0	0/15
Japan, Korea	—	4/0	4/0	0/0	0/0
China	0.00	0/4	0/0	0/0	0/4
Philipphines, SE Asia	0.00	0/12	0/2	0/0	0/10

(1975 who thought MS rates were similar regardless of race in a given locale; their evidence was primarily for Israel.

3.4.4 Familial frequency

The frequency of multiple cases of MS in families within large published series was in the order of 6% with a range of some 2 to 17%. Among these series the frequency of MS among sibs and parents was about 1% and 0.5% respectively. When these percentages were corrected for birth rates and likely survival, a prevalence rate of about 400 per 100 000 for siblings and 200 per 100 000 for parents could be estimated. If one ignored the selection bias of most series, these rates would be about 6–8 and 3–4 times the expectations for the general population in areas considered (Kurtzke, 1965a, 1977). Within Hyllested's (1956 national prevalence survey of Denmark, the prevalence rate for sibs was calculated as 362 per 100 000 and for parents 183 (Kurtzke, 1965a). Thus it is likely that there is truly an increased risk of MS among siblings of the affected; some reservations may still be made as to their parents, though a modest excess does seem probable for them as well. I know of no good data on the *offspring* of MS patients; clinical experience suggests its occurrence there is a rare event.

The explanation for the familial increase again brings up the nature–nurture controversy. As to the genetic aspects, the proponents have largely felt obligated to propose either multifactorial (polygenic) inheritance or autosomal recessive inheritance with reduced penetrance. Unbiased data on twins, which might finally settle the genetic component, are inconclusive. Proponents further indicated that the rarity of conjugal MS (which seems to be about the same as in the general population) was an aspect favouring a genetic influence. This last argument though is not valid if childhood onset of

environmental grounds and, as a general principle, the further affected persons in such clusters diverge from the patterns of simple Mendelian inheritance the greater the chance that an intra-family spread of an environmental agent is the sole explanation for the clustering (Ford, 1978).

Twin studies have also added strength to the genetic argument. In the summary by Kuwert (1977), the risk of developing MS for monozygotic twins is 533 times that of the normal population and, for dizygotic twins, is 259 times. These figures are similar to those found by Mackay and Myrianthopoulos (1966) in their classic MS twin study. The high risk faced by the monozygotic twin (20–25% chance of developing MS) supports the concept of a genetic component which is necessary for disease development.* The fact that 75–80% of identical twins are discordant for MS, however, clearly indicates that this gene, or genes, are not sufficient. Moreover, the much greater risk faced by the dizygotic twin compared with other sibs (who genetically are equidistant from the patient) indicates a major role for an environmental factor. Extensive epidemiological data on the prevalence of MS in static and migrating populations have supported the role played by environment (see Acheson, 1977).

Despite the difficulties in interpreting the genetic basis of MS from family and twin studies, the HLA data do suggest the need for invoking an inherited characteristic in the aetiology of MS. As a consequence, it is now reasonable to suggest that MS is the result of an unusual environmental event affecting a genetically susceptible human host. Moreover, the rapid expansion of knowledge about the role of the MHC in controlling the immune response of many animal species raises the possibility that HLA research will assist significantly in providing both a mechanism for pathogenesis and a likely aetiology of MS.

4.2 THE HLA SYSTEM

The antigens of the HLA (human leucocyte antigen) system are expressed on the surface of most cells throughout the body but are particularly well represented on lymphocytes, the cells used in the laboratory for HLA typing. HLA antigens are coded by a cluster of closely linked genes which reside on the short arm of human chromosome 6 (see Fig. 4.1). Five loci termed A, B, C, D and DR have so far been identified and two of these (HLA-D and -DR) appear to be coding for extremely similar products (DR stands for D related). The system is extremely polymorphic, i.e. in any population there is a large number of alleles for each locus (see Table 4.1). It is for this reason that organ transplantation in man is so difficult since the likelihood of finding unrelated individuals who are HLA identical is extremely remote. Each person has ten HLA genes since he or she has inherited five from each parent. The five genes inherited from one parent control the expression of an antigen for each of the five loci, and this set of antigens is known as a haplotype (from *haplos* meaning

*The reader is also referred to the section by J. F. Kurtzke, page 76, on this point. Ed.

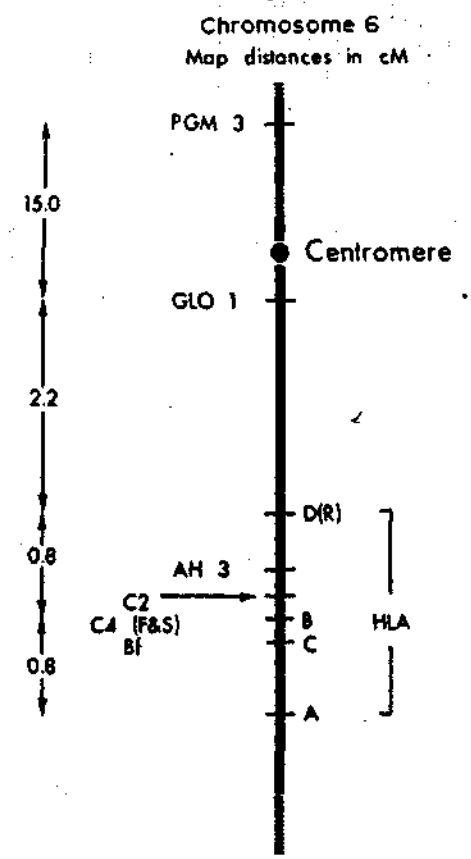

Fig. 4.1 Part of the map of chromosome 6. Based on Francke and Weikamp 1979. PGM = phosphoglucomutase; GLO = glyoxalase; AH = adrenal hyperplasia; C2 and C4 = complement factors 2 and 4; Bf = properdin factor B.

single since these five antigens are coded for by the genes on a single chromosome1. The antigens found on the surface of the cells and reported as the 'HLA type' by the laboratory is known as the phenotype. Haplotype assignment, or genotyping, can only be deduced following typing of several members of a family.

4.2.1 HLA typing

Typing for the HLA A, B and C antigens is carried out serologically using peripheral blood lymphocytes (PBL) or separated T cells.

Typing for the HLA-D locus is performed by mixed lymphocyte culture (MLC). Normally, when the lymphocytes from two unrelated individuals are placed together in tissue culture wells, the cells rapidly proliferate as a result of seeing the foreign HLA-D antigen on each others cells. If one person's lymphocytes are irradiated prior to setting up a culture, the cells are unable

response. Initial concepts were developed from studies of inbred strains of mice and have been valuable pointers for investigations of genetic control of immune responsiveness in man. This mouse research established two major characteristics: the existence of MHC-linked immune response (Ir) genes and the phenomenon of MHC restriction of cellular interactions involving T lymphocytes.

Certain strains of inbred mice are able to respond to particular simple antigens whilst other strains cannot and this ability is controlled by a single dominant 'Ir' gene which resides in the MHC (which in the mouse is known as H-2). More than 30 such genes have been identified as have genes which control immune suppression or Is genes (see McDevitt, 1979). The MHC (H-2) region in the mouse is divided into four sub-regions: K, I, S and D. Sub-regions K and D code for proteins very similar to the HLA A and B antigens and the S region codes for the murine equivalent of the human fourth component of complement (which is coded in man by a locus also in the MHC region). The I sub-region of H-2 was so named since it carries the Ir genes as well as genes controlling the mixed lymphocyte response (the mouse equivalent of HLA-D). It also carries genes coding for surface proteins very similar to the HLA-DR antigens which, in the mouse, are known as Ia antigens. There is some evidence to suggest that the Ia antigens are in fact the mediators of the Ir gene effect, perhaps acting at the level of antigen presentation on the surface of macrophages (Rosenthal, 1978). Since most of the HLA associated diseases are predominantly associated with an allele of the HLA-D (DR) region, it is not surprising that the results of research on Ir genes and Ia antigens in the mouse have caused excitement amongst those interested in the HLA-associated diseases of man.

In the mouse most of the essential cellular interactions involving T cells require that the other cell population and the T cell have common H-2 antigens. This includes the interaction between T cells and macrophages (see Rosenthal, 1978), with B lymphocytes in the production of antibody (see Katz and Benacerraf, 1976), with monocytes in effecting a delayed-type hypersensitivity reaction (Miller and Vadas, 1977) and in the ability of cytotoxic T lymphocytes to lyse virus-infected target cells (Zinkernagel and Doherty, 1974). In addition, the function of suppressor T cells may be MHC controlled (Tada, Taniguchi and David, 1976; see McDevitt, 1979) as may be the intrathymic differentiation of murine T lymphocytes (Zinkernagel *et al.*, 1978).

Similar data in man are much more difficult to obtain. Nevertheless, evidence exists for associations between HLA alleles and altered immune responsiveness to vaccination with tetanus toxoid (Sasazuki *et al.*, 1978), vaccinia (van Rood, De Vries and Munro, 1977) and influenza (Spencer, Cherry and Terasaki, 1976). Other studies supporting the existence of HLA-linked immune response genes include sib-pair analyses of multiple-case families with leprosy (de Vries *et al.*, 1976) and with haemophilia and anti-

factor VIII antibodies (Frommel *et al.*, 1977). This subject has been recently reviewed (van Rood *et al.*, 1977). Evidence for HLA restriction was reported by McMichael *et al.*, 1977) who demonstrated the need for sharing an HLA-A or B locus specificity between influenza-infected target cells and cytotoxic T cells for immune lysis to occur. Sharing of an HLA-D locus antigen between antigen-presenting monocytes and proliferating T cells was required for optimal stimulation by purified protein derivative (PPD) in one study (Bergholtz and Thorsby, 1978). Anti HLA-D antibodies have recently been shown to block T lymphocyte transformation in response to several antigens (Bergholtz and Thorsby, 1978; Greenberg *et al.*, 1978; Breard *et al.*, 1979; Stewart *et al.*, 1981a) suggesting that human macrophages present antigen to T cells in combination with an HLA-D locus product.

The HLA system also holds particular appeal for geneticists in view of two phenomena: the uniquely high polymorphism of the system (see Table 4.1) and the existence of marked linkage disequilibria. Since it is likely that the immune system recognizes viruses (and other intracellular infectious organisms) in combination with HLA antigens on the cell surface (altered self hypothesis), the high degree of polymorphism may have evolved in response to new pathogens which mimic MHC products and thereby escape immune detection (Bodmer and Bodmer, 1978).

The other phenomenon, linkage disequilibrium is defined as the association at a population level of alleles of two linked loci more frequently than would be expected by chance. For example, HLA-B8 and Dw3 or HLA-B7 and Dw2 occur together with a much higher frequency than expected. This has been interpreted to suggest that products of more than one locus within the HLA system selectively interact providing a specific advantage to the individual who keeps these antigens together (Bodmer and Thomson, 1977). Alternatively, it may be due simply to the fact that the gene loci are so close together there has not been enough time for the genes in question to be mixed randomly by the process of chromosomal recombination. Whichever explanation is correct, the concept of linkage disequilibrium is central to understanding HLA and disease associations. It is likely that the HLA gene itself is not the disease susceptibility gene (DSG) but is merely a marker for a closely linked DSG (see Fig. 4.2) with which it is in linkage disequilibrium. This complex subject has been well reviewed (Bodmer and Thomson, 1977; Bodmer and Bodmer, 1978).

4.4 MS POPULATION STUDIES

The first reports of an association between multiple sclerosis and the HLA system appeared in the early 1970s (Bertrams, Kuwert and Liedtke, 1972; Naito *et al.*, 1972; Jersild *et al.*, 1973a). By 1975, Jersild *et al.* were able to summarize data from several centres in both North America and Europe indicating definite associations between MS and HLA-A3 and HLA-B7. In the same review, they presented evidence from Copenhagen, Munich and Oslo on

CHAPTER FIVE

Clinical Aspects of Multiple Sclerosis

J. F. Hallpike

5.1 Introduction
5.2 Historical aspects
5.3 Classification
5.4 Symptomatology
5.5 Natural history and prognosis
5.6 Diagnosis
5.7 Differential diagnosis
5.8 Conclusions

5.1 INTRODUCTION

What is multiple sclerosis (MS)? What are the clinical features of the disease? How is the diagnosis established? What is the differential diagnosis? What is the natural history and prognosis of MS? Discussion of these questions forms the theme of this chapter. The spectrum of symptoms, signs and course of MS is characteristically varied. On the other hand, the pathological changes in the central nervous system (CNS) are remarkably consistent and MS presents, therefore, as a distinctive clinicopathological entity. Nevertheless, in the absence of a definitive understanding of the cause or pathogenesis of MS it has not been possible to devise a laboratory test which is reliable and diagnostically specific. Physical examination alone rarely yields findings which are pathognomonic of MS. In clinical terms, the disease appears as a profile or syndrome in which history and CNS signs are the main features but in which other factors such as local MS prevalence, early residence data, genetic susceptibility also have a bearing on diagnostic confidence and accuracy. Evoked response studies complement the physical examination by providing evidence of subclinical conduction delays in CNS pathways and may also have a role in monitoring disease progression. In the correct clinical context, cerebrospinal fluid (CSF) examination is frequently informative and findings such as a moderate mononuclear pleocytosis or evidence of synthesis of oligoclonal IgG within the blood brain barrier (BBB) are important indications of MS. Computerized tomography (CT) is capable of revealing white matter changes in MS brain. Very recent application of monoclonal antibody techniques in MS suggests that T cell subset ratios in peripheral blood may have a place in diagnosis and follow up. These investigative

approaches are modifying traditional views about MS. Laboratory data indicative of MS are replacing as well as complementing clinical signs of CNS disease for diagnostic purposes. Hence, in clinical practice the spectrum of early or milder forms of the disease has widened with an increased emphasis being placed on investigative findings. The implications of this trend for any new view of the natural history of MS are not yet clear. Nevertheless, clinical evaluation remains extremely important in the diagnostic process and for the assessment of disability. Early diagnosis places an added burden on the physician who can expect to be intensively quizzed about the relevance and prognostic significance of symptoms, certainty of the diagnosis and the value and availability of putative treatments.

5.2 HISTORICAL ASPECTS

The first clinical and pathological descriptions of MS (disseminated sclerosis, sclérose en plaques) were published a little over a century ago. In the ensuing years, this condition has probably been the subject of more investigation and speculation than any other disease of the nervous system. MS is one of the commonest organic neurological disorders and the most frequently encountered human demyelinating disease. Onset characteristically is in early adult life and, although the course is variable and sometimes benign, progression is usual so that in parts of the world where MS is prevalent (see Kurtzke, Chapter 3) the illness may be compared with stroke and trauma as a cause of socio-economic hardship. Credit for the earliest clinicopathological descriptions of MS is generally accorded to Jean Cruveilhier (1835–1842) and Robert Carswell (1838). The first comprehensive account of MS was by Charcot (1877) who identified key clinical features such as dysarthric speech, ataxia, tremor, paresis and visual defects and distinguished this syndrome from other chronic CNS disorders such as tabes dorsalis and paralysis agitans. Further contributions of that period and the turn of the century by Frerichs, Rokitansky, Rindfleish, Vulpian, Marburg and others are reviewed by Dejong (1970). Early English reports were those of Moxon (1873) and Gowers (1888). Seguin (1880) provided one of the first American accounts which also served to emphasize the common spino-optic form of the condition. Müller's monograph (Müller, 1904) contained 1100 references. The papers of Dawson (1916) were a landmark in the histopathology of MS and the review by Brain (1930) included a highly pertinent discussion of the relationship between MS and other demyelinating diseases. More recent major sources of information are the McAlpine books (1955, 1972 and the *British Medical Bulletin* (Porterfield, 1977) devoted to MS.

Many factors have at different times been held to play a part in the aetiology of MS. This diverse and intriguing evidence cannot yet be assembled into a

fully coherent picture. Nevertheless, epidemiological evidence, genetically influenced 'susceptibility' and immunological considerations are paramount in current attempts to devise a model of the MS disease process. Systematic epidemiological studies of MS begun in the early 1950s have delineated high and low prevalence regions within which certain ethnic groups appear to be more or less liable to develop the disease. The effect of age of migration from high to low risk zones on subsequent occurrence of MS is one of the strongest arguments favouring the operation of an environmental pathogenetic agent. Nevertheless, persistent attempts to isolate virus from tissues of affected patients have conspicuously failed to provide reproducible evidence of an MS-specific viral agent. There is a tendency for patients with MS to have higher titres of antibodies in CSF and serum to a range of common viruses, particularly measles, than comparable controls (see ter Meulen, Chapter 9). The inconstant and variable nature of these findings makes their direct relevance to plaque pathogenesis improbable. However, it does appear that MS patients as a group exhibit an impairment of cell-mediated immunity currently most specifically manifested as a defect of peripheral suppressor cell activity (Johnson, 1980). The possibility of a vascular cause for MS has a long history (see Adams, 1972) but venule occlusion in plaques is no longer thought to be a pathogenetic event. It has been suggested that a failure of oligodendrocytes could be responsible for loss of myelin and, in general, hypotheses implicating glia in the pathogenesis of plaques remain attractive in view of the important nutritive and stabilizing functions of many of these cells (Lumsden, 1970a). Search for a primary lipid abnormality predisposing to demyelination has been a longstanding topic of research in MS. There is no evidence that MS is due to a genetic defect of myelin lipids. Certain data concerning polyunsaturated fatty acids in MS are discussed elsewhere (Hallpike, Chapter 11) but have not so far been shown to be disease-specific. The concept of MS as an autoimmune disease originated with the work of Rivers (Rivers and Schwentker, 1935) and Kabat (Kabat, Wolf and Bezer, 1946) on the production of experimental allergic encephalomyelitis (EAE) by inoculations of nervous tissue. Encephalitogenicity was found to depend on the basic protein (MBP) component of myelin (Einstein *et al.*, 1962) and arguments for and against the validity of the EAE model of MS are dealt with in detail elsewhere in this book but have been usefully summarized by Carnegie and Moore (1980).

Much of the basic work on demyelinating diseases in the last 20 years has stemmed from a special interest in myelin as a highly distinctive membrane system. The mechanisms of normal production and maintenance of myelin are germane to the demyelinating process (Gregson, Chapter 1). It is possible that effective treatment of MS will antedate a solution of the aetiopathogenesis of the disease. However, the inherent difficulties in proving the worth of putative therapeutic regimens by controlled clinical trials are very great. The overriding requirement is for continued systematic application of a basic

circumscribed nature of the CNS lesions. The view that brain oedema could cause demyelination has not been substantiated in experimental studies (Ibrahim, Morgan and Adams, 1965; Adams, 1965) and selective myelin loss does not occur in Reye's syndrome, following head injury or adjacent to cerebral neoplasms.

5.3.2 Multiple sclerosis

Until a specific and reliable test becomes available, the diagnosis of MS will continue to rely mainly on clinical criteria. Proof of diagnosis ultimately rests with autopsy confirmation—'MS proved by autopsy'. The clinician, however, and most investigators require a set of criteria, applicable in life, to permit classification which reflects degree of certainty of diagnosis and which underlines the probability aspect of the assessment. Any such clinical classification should be reasonably straightforward and designed in the interests of uniformity of diagnostic standards and world-wide application. The fact that MS is both an uncommon and a very variable disease means that, for instance, epidemiological surveys and therapeutic trials have generally to be conducted on a multicentric basis involving numbers of data gatherers: findings have to be independently tested so that case-selection and evaluation according to a generally accepted scheme appears to be essential. The next and very topical question that arises in this context of MS diagnosis is the importance to be attached to laboratory findings. Is it appropriate to assimilate evoked response and CSF results into present clinical classifications? While it is obvious that CSF and neurophysiological studies are vital to scientific understanding of MS, such tests have not been shown to predict the course of the disease. Hence, the relevance of subclinical CNS disease, demonstrated by these means, to disability and the likelihood of progression has yet to be defined. Abnormal laboratory profiles provide evidence of multifocal or more general subclinical disease which can be of great value in managing early or suspected cases and obviating requirements for invasive radiology. Nevertheless, from the viewpoint of patient and doctor the main watershed in diagnosis is between clinically definite on the one hand and probable or possible categories (Table 5.2) on the other. There are practical implications of a definite diagnosis of MS which can be helpfully and constructively discussed. Confusion arising from tentative diagnosis, sometimes supported by an abnormal evoked response study or a raised CSF IgG, can be distressing and a cause of unnecessary gloom. In the event of effective therapy for MS becoming available, there will be a much greater premium on early recognition of disease short of 'clinical certainty'. In that event, laboratory data would be expected to figure prominently in diagnostic guidelines devised empirically for treatment purposes.

In the 1975 Revision of International Classification of Diseases WHO, 1977), multiple sclerosis (340) appears under 'Other Disorders of the Nervous

Table 5.2 Clinical diagnosis of multiple sclerosis

Clinically definite MS
Fulfilling Schumacher criteria

Probable multiple sclerosis
1. Relapsing/remitting symptoms with only one neurologic sign commonly associated with MS; or
2. Documented single episode with signs of multifocal white matter disease with complete or partial recovery
3. No better explanation

Possible multiple sclerosis
1. Relapsing/remitting symptoms without documented signs; or
2. Objective signs insufficient to establish more than one site of CNS involvement
3. No better explanation

Rose *et al.* (1976).

System' with brainstem, cord and generalized types. Certain rarer demyelinating diseases of the CNS (341) have individual subheadings, i.e. neuromyelitis optica (341.0), Schilder's disease (341.1) and so on. This WHO classification tends to be confusing because of additional listings for disabilities, such as hemiplegia, quadriplegia, irrespective of causation. The definition of MS proposed by the Schumacher Committee (Schumacher *et al.*, 1965) is still the most widely used. The six criteria, linking natural history with physical findings, considered essential for 'clinically definite multiple sclerosis' are shown in Table 5.3. The Committee emphasized that diagnosis, even in the 'clinically definite' category, remains one of high probability only, because of the absence of a specific diagnostic test. The problem of providing a wider spectrum of diagnostic probability than offered by the Schumacher criteria has been approached in various ways. Clinically 'definite', 'probable' and 'possible' groups were defined by McAlpine (1972) and are closely resembled by the more formal proposal of Rose *et al.* (1976) in which 'definite' satisfies

Table 5.3 Clinically definite MS—Schumacher criteria

1. Neurological examination reveals objective abnormalities of CNS function
2. Examination or history indicates involvement of two or more parts of CNS
3. CNS disease predominantly reflects white matter involvement
4. Involvement of CNS follows 1 or 2 patterns:
   i. Two or more episodes, each lasting at least 24 h and a month or more apart
   ii. Slow or stepwise progression of signs and symptoms over at least 6 months
5. Patient 10—50 years old at onset
6. Signs and symptoms cannot better be explained by other disease process

Schumacher *et al.* (1965).

the Schumacher criteria and 'probable' and 'possible' reflect lesser degrees of CNS involvement or evidence of progression (Table 5.2). The classification described by McDonald and Halliday (1977) is more comprehensive with 'clinically definite', 'early probable or latent', 'progressive probable', 'progressive possible', 'suspected' and 'necropsy-proved' categories. The criteria are clinical with the addition that electrophysiological testing may also be employed to provide objective evidence of lesions. Such a detailed scheme has the merit of an input from an important methodology and should permit patients to be more accurately grouped for comparison purposes with respect to prospective studies of natural history or treatment. A numerical scoring system has been devised for classification (Poser, 1979) which enables incorporation of data from evoked potential studies, psychological testing and other methods for delineating asymptomatic lesions. Another attempt to integrate laboratory findings into the grading of diagnosis set out in the Rose scheme has been made by Bauer (1980) on the basis of a survey of opinion of neurologists and MS scientists. Strong support was recorded for standards for MS diagnosis based on clinical features, course and laboratory findings. Although a majority of those surveyed favoured inclusion of first bouts under a diagnosis of MS if disseminated symptomatology and characteristic CSF findings were present, an important minority opinion was opposed to designating first bout cases as 'clinically definite'. Other conclusions were to make characteristic CSF findings an obligatory criterion for the 'clinically definite' group and to locate monosymptomatic retrobulbar neuritis in the 'clinically possible MS' group. For routine purposes it appears that the Rose classification, with the addition of 'CSF profile indicative of MS' as a requirement in the 'clinically definite' group, most closely reflects present views. More complex schemes may be appropriate for particular projects or trials and it continues to be important to specify the use of evoked potential results.

In addition to classification of MS with respect to diagnostic probability, there are also distinct and well-recognized categories of 'clinically definite' disease. In relapsing and remitting MS there is a fluctuating course with clear-cut relapses and recovery and only minimal or moderate disability between relapses. Chronic progressive MS is a phase in which disability is steadily progressing: a further common pattern is 'relapse-remitting becoming progressive'. Benign MS is the term applied where disease has been present for 15 years or more and the score on the Kurtzke Disability Status Scale (Section 5.4.3) is not more than 3 at time of examination.

5.4 SYMPTOMATOLOGY

The clinical features of MS are protean because plaque formation is ubiquitous and no myelinated connecting system is exempt from attack. Reviews of the symptomatology of MS can be daunting in view of the many

permutations of symptoms and signs that occur and difficulty picking out themes of special interest. Nevertheless, as already emphasized, an understanding of the clinical aspects of the disease is essential for diagnosis and management and is still the reference point for most research. Within the CNS, there are sites of predilection for MS resulting in a number of more commonly encountered patterns of CNS dysfunction. The clinical profile of MS involves a 'cross sectional' account of symptoms and findings at a time of examination coupled with specification of the 'longitudinal' course of the illness. The purpose of clinical examination may be considered to be three-fold: elicitation of criteria for diagnostic decision making, accurate documentation for the record and as a basis for research and, thirdly, practical assessment of disability. Difficulties can be experienced translating the findings of a traditional neurological examination into a clear picture of disablement. Use of disability scales is, therefore, an important requirement particularly in relation to judging therapy and for the provision of aids and services. The discussion of symptoms and signs of MS which follows is under three headings: major or common features, unusual features and disability assessment.

5.4.1 Major features

The onset of symptoms in MS is extremely variable ranging from acute or subacute to chronic progression of disability over months or years. The 'onset' of MS refers to the occurrence and timing of the first symptom or deficit that can definitely be attributed to the disease. The term 'relapse' describes the development of fresh symptoms and 'remission' connotes a complete or partial resolution. As noted in the Schumacher criteria (Table 5.3) MS rarely commences in childhood or over the age of 50. MS is, however, not infrequently diagnosed in the 50–60 age group where the history suggests longstanding disease activity and appropriate laboratory results are obtained. In one series (McAlpine, 1972) onset was monosymptomatic in 45% of 241 cases. The percentage of cases with a single deficit onset was considerably less in the large US veterans series (Kurtzke *et al.*, 1968) but, as a group, these patients appeared to have a better prognosis than those with multiple involvements in the initial bout. A breakdown of overall symptoms in MS taken from a study of 810 patients (Müller, 1949) is shown in Table 5.4. Disturbances of balance are the most frequently encountered class of symptoms, followed by sensory loss, weakness and micturition impairment. Poor balance is, in fact, a composite complaint usually due to weakness or cerebellar or sensory changes as well as brainstem, auditory or even visual involvement. Motor symptoms are probably the single commonest cause of disability in MS. Common early complaints are of heaviness or dragging of a leg, of undue fatigue or of tripping. Even if leg symptoms are unilateral, signs of corticospinal or pyramidal tract involvement are commonly bilateral at an

Table 5.4 Frequency of symptoms in MS

Balance abnormalities	78%	Paraesthesiae	40%
Impaired sensation	71%	Giddiness	32%
Paraparesis	62%	Hemiparesis	18%
Micturition changes	62%	Facial palsy	15%
Optic neuritis	55%	Epilepsy	5%
Monoparesis	52%	Impotence	5%
Ataxia of limbs	45%	Hearing loss	4%
Diplopia	43%	Tic douloureux	2%

Müller (1949).

early stage. Leg weakness in MS usually reflects spinal cord disease although the evolution of spastic paraparesis or quadriparesis is often asymmetrical. Hand weakness is manifested as loss of dexterity affecting handwriting and manipulative skills: profound upper limb paresis is much less frequently seen than severe paraparesis or paraplegia. A hemiparetic distribution of weakness including the face occurs in MS but careful clinical examination will generally yield signs of bilateral disease, i.e. both plantar responses extensor. Slow evolution of hemiparesis without contralateral or disseminated signs is always more suspicious of a space-occupying lesion such as a meningioma or low-grade astrocytoma. Pathological exaggeration of the jaw jerk is a valuable sign of pyramidal disease above pontine level: loss of abdominal reflexes, particularly in younger non-obese subjects, appears as a subtle indication of corticospinal tract involvement in MS. Gait disturbance in MS, sometimes represented initially as unsteadiness, is the most frequent effect of pyramidal tract dysfunction. Focal muscular wasting is rarely found in MS. An exception to this is lower motor neurone facial paresis which can be an early, often isolated, manifestation of the disease readily confused with the common idiopathic Bell's palsy. In the limbs, hand muscle atrophy seems to occur occasionally and may be due to impingement of sclerotic plaques on anterior horn cells or demyelination of motor efferents within the cord. The question of a rare association between MS and generalized radiculopathy or peripheral neuropathy is referred to briefly below (5.4.2). In general, localized neurogenic wasting is most often due to decubital peripheral nerve compression

Impaired sensory functions and paraesthesiae together comprise another major group of symptoms in MS. Involvement of the visual pathways due to optic neuritis (ON) is perhaps the most clearly defined of all the sensory changes in MS and is considered fully elsewhere in the book (Moore, Chapter 6). Paraesthesiae, i.e. tinglings, pins and needles and other 'positive' sensory symptoms reflect interference with conduction in sensory pathways at any level. Disturbances of posterior column modalities, e.g. vibration and joint

position sense or two point discrimination, are the commonest sensory deficits found in spinal cord disease. In addition to sensory ataxia and Rombergism, symptoms of posterior column disease may include severe impairment of manipulative and discriminative functions of the fingers, 'tightness', bandage sensations and numbness. Lesions involving root-entry zones can also give rise to tingling, hyperpathia or, rarely, pain in girdle or segmental distributions. Lhermitte's sign, an 'electric shock' sensation in the back and limbs provoked by sharp neck flexion, is best regarded as evidence of organic disease of sensory pathways in the cervical cord but not as being indicative of or peculiar to MS. Lhermitte's sign is also seen occasionally with extramedullary compression and in subacute combined degeneration of the cord (Gautier-Smith, 1973). Superficial sensory loss is much less frequent in MS than the changes associated with posterior column lesions. Pin-prick and temperature impairment due to spinothalamic involvement tends to be unilateral and is often part of a partial or complete hemi-cord or Brown-Séquard syndrome. Light touch appreciation is preserved to a surprising extent in MS even in the presence of very gross disability. Global sensory loss affecting a limb or limbs or from the neck down raises a strong suspicion about organicity. Certain unusual but distinctive sensory manifestations, namely trigeminal neuralgia and other episodes of a paroxysmal nature are considered separately (5.4.2). A chronic pain syndrome associated with altered spinothalamic sensation is a rare occurrence in MS.

Evidence of cerebellar system involvement is common in MS. Charcot's triad of dysarthria, nystagmus and intention tremor was early recognition of the importance of cerebellar signs in MS although the triad itself is usually only found in more advanced disease. The most frequent type of cerebellar abnormality in MS is an ataxia of stance and gait which, when combined with a spastic paraparesis, results in a distinctive pattern of cerebellar and pyramidal involvement of 'spastic–ataxic' gait. Nystagmus is an important sign of cerebellar system disease but, of course, also commonly results from vestibular and intrisic brainstem lesions as well as from extra-axial compressions. Modern oculographic methods permit a more accurate approach to classifying nystagmus. However, the clinician may still find it hard to identify the anatomical lesion unless a specific abnormal pattern is present, e.g. an internuclear ophthalmoplegia, seesaw nystagmus etc. (see Daroff and Dell'Osso, 1979). According to Kurtzke (1970 cerebellar abnormalities were present in 70% of a veterans series although a considerably lower figure is given by McAlpine (1972). Cerebellar system disease would contribute substantially to 3 of the more common types of disability listed in Table 5.4.

In addition to involving cerebellar pathways, brainstem lesions in MS affect the long tracts and local mesencephalic, pontine and medullary connections. Monofocal brainstem presentations in the younger age group can readily be misdiagnosed as MS if the principles underlying the Schumacher criteria are

not applied. Unless the course of such types of disease, evoked potential and CSF findings, conform to the MS pattern, persisting localized brainstem lesions should arouse suspicion of tumour. External ocular palsies occur in about 30% of cases at some time during the illness (Kahana, Leibowitz and Alter, 1973a). The neuro-ophthalmological features of MS are considered elsewhere (Moore, Chapter 6). The effects of widespread brainstem involvement often reflect the level at which plaques are concentrated. Thus, extensive mesencephalic demyelination can produce a picture of mutism, ophthalmoplegia, pseudobulbar changes and bilateral long tract signs. Pontine disease most frequently leads to pareses of conjugate lateral gaze, trigeminal signs, facial weakness, pseudobulbar palsy and varying degrees of bilateral limb weakness, ataxia and sensory impairment. An acute medullary syndrome is seen rarely in MS but is one of the major causes of very early death in this disease due to involvement of reticular and autonomic regulating systems, bulbar palsy and quadriplegia.

Some degree of impairment of control of bladder function is a very frequent occurrence in the course of MS (Table 5.4; Kurtzke, 1970; Bradley, 1978). Symptoms, ranging from mild hesitancy or urgency to retention or incontinence, usually bear a relationship to overall disability. Presentation with a marked persistent micturition disturbance is unusual. Disinhibited detrusor activity with a small capacity bladder, readily exacerbated by lower urinary tract infection, is the commonest pattern of dysfunction: the problem is reviewed in much greater detail elsewhere (Parsons, Chapter 17). Impotence is another important category of autonomic disturbance in MS with an incidence of symptoms ranging from 5–80% in different series (Kurtzke, 1970; Vas, 1969; Lilius, Valtonen and Wikström, 1976). Worsening of symptoms and signs in MS with elevations of body temperature has long been recognized with respect to visual acuity and field defects in optic neuritis (Uhthoff's phenomenon). This effect is now explained in general terms through an understanding of temperature effects on partially demyelinated fibres, whereby temperature increases of as little as 0.5°C can cause reversible conduction block (Rasminsky, 1973). Hyperthermia testing has been employed in various ways as a diagnostic test in MS and to reveal latent abnormalities (Davis, Michael and Neer, 1973; Jestico and Ellis, 1978; Bajada *et al.*, 1980). Certain investigations of visual function employing a temperature change, for instance double-flash perception and critical flicker fusion frequency, appear to be very sensitive methods for detecting subtle conduction abnormalities in the visual pathways (Regan, 1979). An implication of these and other neurophysiological studies in MS (see Sedgwick, Chapter 7) is that subclinical disease is extremely common. It has been strongly suggested that randomly distributed lesions in the peripheral nervous system are a basis for the proximodistal gradient of sensory impairment, found in peripheral neuropathy (Waxman *et al.*, 1976). Such an approach, invoking summation effects of lesions, may also help to explain the

common occurrence in MS of such changes as slowly progressive paraparesis, frequent bladder involvement and ataxia in later stages of the disease.

5.4.2 Unusual features

The occurrence of *dementia* in MS is probably under-recognized as the clinical picture is usually dominated by more obvious disabilities. Performance in routine tests of intellectual function is often passable, but systematic psychometric evaluation has suggested that some degree of cognitive impairment may be present in up to 50% of MS patients (Peyser *et al.*, 1980). Overt organic dementia is an uncommon presenting feature of MS (Bergin, 1957; Koenig, 1968) but is present with widespread cerebral involvement. The 'euphoria' often referred to in this disease correlates well with intellectual deterioration and denial of illness (Surridge, 1969). *Psychotic* manifestations are also rare. Occasionally, however, schizophrenic symptoms are found co-existent with MS (Parker, 1956; Hollender and Steckler, 1972) and Matthews (1979) has recently drawn attention to the importance of considering MS as a cause of acute remitting mental symptoms in young adults. For the most part, psychiatric symptomatology in MS is clearly secondary to the disease but depression does appear to be so common that it has been suggested (Whitlock and Siskind, 1980) that serious affective disorder might be a presenting feature. No association was found between MS and premorbid personality types (Pratt, 1951).

Kinnier Wilson and Macbride (1925) gave an early account of the association of MS with *epilepsy* and pointed to the occurrence of Jacksonian or partial motor seizures. Estimates of the true incidence of epilepsy in MS vary but a figure of about 2% would appear to be reasonably accurate (Matthews, 1962; McAlpine, 1972) although a higher figure is given in Table 5.4 taken from Müller's data. Seizures, which are usually infrequent, can be both focal and generalized and occur in the course of the disease rather than at the outset. *Trigeminal neuralgia* indistinguishable from tic douloureux appears to occur in about 2% of patients with MS. Paroxysmal trigeminal tic occurring in MS may not be preceded by facial numbness but is more frequently bilateral than the classical idiopathic condition (Harris, 1950). In Peet and Schneider's (1952) survey of 689 cases of trigeminal neuralgia, MS was present in only 8 or just over 1% of the patients. The question of whether tic is a true symptom of MS or a chance association was more specifically considered by Rushton and Olafson (1965) in their review of the Mayo Clinic experience. Out of 5615 cases of trigeminal neuralgia and MS seen at the Clinic between 1948–62, 35 were identified with both conditions. Thus, there was no substantial evidence of a cause and effect relationship and it was noted that, in general, organic disease of the descending root of the trigeminal nerve caused sensory impairment rather than tic. Glossopharyngeal neuralgia was not encountered

as a symptom of MS in the Mayo Clinic series. A peripheral type of *facial palsy* occurs sometimes as an early feature of MS and, in the absence of other signs of neurological disease, will often be thought initially to be a Bell's palsy. Facial weakness in MS may also be central as part of a pontine or hemiparetic syndrome. The incidence of facial palsy in MS is variously quoted from 2% (Kurtzke, 1970) to 15% (Müller, Table 5.4). The condition of *facial myokymia*, clearly described by Matthews (1966), comprises a state of unilateral involuntary rapid twitching or flickering of facial muscles without appreciable weakness. Duration of symptoms is usually from a few weeks to several months. Although there appears to be a highly significant association of facial myokymia with MS, the finding may also be due to other intramedullary disease.

Paroxysmal disturbances occur infrequently in MS and take the form mainly of short-lived, painful, motor and sensory phenomena arising focally within the brainstem or cervical cord (Matthews, 1975; Perks and Lascelles, 1976; Twomey and Espir, 1980). Attacks of transient dysarthria, tonic head turning and paroxysmal sensory symptoms are aspects of this syndrome. Precipitation by exertion or hot bath supports the view that these events reflect aberrant conduction ephaptic in partially demyelinated fibres (Osterman and Westberg, 1975). In a recent personal case, a 52-year-old woman was referred for investigation of frequent episodes of transient unilateral limb weakness, mimicking TIAs, and was shown conclusively to be suffering from MS without coincident major vessel disease. Her attacks ceased with carbamazepine, which is commonly an effective treatment of paroxysmal episodes in MS. It is likely that with improved diagnostic methods and increased awareness of the diversity of symptoms in MS, atypical presentations of a quasi vascular or epileptic nature will more often be shown to be due to demyelinating disease.

Changes in the appearances of the retinal veins in MS are of considerable interest, being referred to as '*retinal periphlebitis*' or perivenous sheathing. Mild retinal perivenous sheathing appears to have an incidence of about 1% in MS (Rucker, 1944; Bamford *et al.*, 1978). Uveitis is a rarer association, while the condition of primary retinal haemorrhages in young males (Eales' disease) does not appear to have any relationship to MS (Hörnstein, 1971). The presence of ophthalmoscopic abnormalities of veins in MS provides support for the suggestion (Fog, 1965) that plaques in the CNS are associated with perivenular inflammatory changes. Another rare association with MS which has just been reviewed is the occurrence of cerebral *glioma* (Anderson *et al.*, 1980). Current opinion tends to favour the possibility of neoplastic transformation of glia in areas of established demyelination in MS, although a chance association of the two conditions cannot be excluded. An uncommon finding in MS is the occurrence of unilateral *deafness*, due to involvement of peripheral cochlear neurones in the neuroglial part of their course (Dix, 1965). Bilateral deafness is also seen with brainstem lesions, including MS, rostral to the cochlear nuclei and has been studied with

particular reference to the question of central recruitment of loudness (Dix and Hood, 1973).

Peripheral nerve abnormalities are not a feature of the usual clinicopathological profile of MS. Plaque formation is confined to the CNS in MS, suggesting that immunological differences between CNS and PNS myelin as well as the presence of glia may be relevant to pathogenesis. Nevertheless, recent studies of the peripheral nervous system in MS indicate that there may be some association with peripheral neuropathy. Sural nerve biopsies from MS patients with little disability showed mild demyelinating changes (Pollock, Calder and Allpress. 1977). Associations of MS with hypertrophic polyneuropathy (Schoene *et al.*, 1977) and with acute demyelinating polyradiculoneuropathy (Forrester and Lascelles, 1979; Lassmann, Budka and Schnaberth, 1981) have also been documented. The hypertrophic neuropathy study was of particular interest in that distinctive 'onion bulb' formations were also found in the CNS within MS plaques. Autonomic disturbances are relatively common in MS and for the most part are central in nature. Additional to abnormalities of bladder function, considered elsewhere (Parsons, Chapter 17 . more severely disabled MS patients commonly display defective thermoregulatory sweating without associated orthostatic hypotension (Cartlidge. 1972). *Respiratory failure* is rare in MS and usually reflects extensive bulbar disease. Reversible respiratory paralysis has been described due to localized medullary demyelination (Boor *et al.*, 1977). A more general account of central neurogenic respiratory dysfunction patterns is given by Plum and Alvord (1964).

Although MS occurs rarely under the age of 15 years, there are reports of young children with relapse-remitting CNS illness and abnormal CSF profiles typical of patterns encountered in the adult condition. The matter of *paediatric MS* is of principal importance because of the current view that the usual development of MS follows exposure to an exogenous risk factor during childhood (Kurtzke, Chapter 3). The Mayo Clinic experience appears to be the most extensive with an account of MS in 40 children aged 7–14 years at onset (Gall *et al.*, 1958). In this series, twice as many girls were affected as boys and a rather high proportion of cases became severely disabled or had died by the late teens. There are other well documented case reports of paediatric MS (Low and Carter, 1956; Isler, 1961; Kuroiwa *et al.*, 1962). A follow-up of 30 children presenting with optic neuritis between the ages of 5 and 15 years showed that 8 of the group went on to develop MS 2.5–16 years later (Kennedy and Carter. 1961). A psychotic presentation of MS in an 11-year-old girl with autopsy confirmation has also been described (Salguero, Itabashi and Gutierrez, 1969 .

5.4.3 Disability profiles

Disability assessment in MS is a problem in its own right which is also

considered in relation to rehabilitation (Colville, Chapter 20). Clinical neurological examination is strongly orientated towards the anatomical location of lesions and establishing a diagnosis. Although a correlation exists between abnormal findings and disability, this relationship is not straightforward and many key signs, for example the extensor plantar response, optic atrophy, spontaneous nystagmus, have no direct bearing on disability. Thus, a well-documented neurological examination which is excellent for purposes of diagnosis can, nevertheless, be singularly uninformative about incapacities for daily living. Failure to appreciate the different objectives of examination for diagnostic purposes on the one hand and accurate disability assessment on the other is still a source of misunderstanding between the physician, therapists and patients. It appears to the writer that the traditional commitment of the neurologist to a diagnostic role is not always paralleled by a corresponding interest in disability scaling which is the current yardstick for therapy and management. The architect of the most widely adopted approach to this problem is Kurtzke (1955, 1961, 1965). The Kurtzke scales as applied to systems (pyramidal, cerebellar, brainstem, sensory, bowel and bladder, visual and mental) are biased strongly towards function deficits and are complemented by an overall Disability Status Scale (DSS) concerned with activities of daily living. This DSS and systems scoring, with minor local modifications, are the most widely employed means of assessing disability in MS, particularly in studies of natural history of the disease and objectively evaluating putative therapies. A disadvantage of the Kurtzke systems scales is that they are designed to be administered by someone with knowledge of clinical neurology. Although the DSS overcomes this, differences between some grades are insufficiently refined to permit accounting of significant functional changes occurring in the course of individual patients. Other approaches to disability assessment have more openly acknowledged the poor correlation between physical signs and disablement and have focused on description and testing of daily activity levels as well as facilitation of record keeping by non-medical personnel (Pedersen, 1965; Potvin and Tourtellotte, 1975). There can be no simple answer to the problem of quantitative analysis of disability in a disease as complicated and variable as MS. The Kurtzke scales are widely employed and understood by those who conduct clinical trials or engage in epidemiology and can be complemented by study of additional variables as appropriate. There is a natural interest in the place of laboratory data in relation to disability in MS. Currently there is no reliable laboratory index of disease activity although some studies (Olsson, Link and Müller, 1976; Link *et al.*, 1977; Tourtellotte, Chapters 10 and 16) suggest that CSF IgG levels are most frequently and significantly elevated in more disabled patients. Abnormal evoked responses are a good guide to the siting of lesions and presence of subclinical disease but are a poor indication of disability. Quantitative EMG techniques can, however, be devised to measure motor deficits in MS (Benecke and Conrad, 1980). For most purposes disability

assessment is best considered in clinical terms with emphasis on the matters of chief concern to the patient.

5.5 NATURAL HISTORY AND PROGNOSIS

The pattern of MS can be specified in terms of *clinical type* and *clinical course*. Commonly designated types are disseminated, spinal, spinal and optic, spinocerebellar and brainstem. The course of MS may be benign, exacerbating–remitting, progressive from outset or exacerbating–remitting becoming progressive. A particular problem arises in relation to isolated optic neuritis (ON) which some would see as a manifestation of MS but which is hard to classify as such in the absence of any other clinical evidence. Consideration of the natural history involves discussion of mortality data on the one hand and on the other recognition that MS may remain clinically silent to be found only as an incidental autopsy finding. Practical interest centres particularly on attempts to define adverse clinical patterns early in the disease, general statements about the probability of long-term disability and identification of possible aggravating factors. Few diseases offer greater difficulties in prognosis than MS (Brain, 1936).

The age of onset of MS derived from several series is best expressed as being between 20 and 40 in approximately 70% of cases (Kurtzke, 1970). The corresponding figure from McAlpine (1972) for this age bracket is 72% for males and 71% for females and a figure of precisely 70% is given by Confavreux, Aimard, and Devic (1980). A slight female preponderance of about 6:4 is a feature of many series but is not invariable. Most surveys have not shown a correlation between age of onset and long-term disability although at least one report (Leibowitz, Kahana and Alter, 1969) suggests that a late onset is associated with a poorer outlook. Estimates of the frequency of exacerbations and duration of remissions have varied through lack of uniformity concerning definition of these phases and because of the tendency for clearly defined 'bouts' to merge with the chronic progressive stage of the disease. Initial symptoms, early frequency of relapses, age and laboratory findings at diagnosis bore no relationship to the later course of the illness (Kurtzke *et al.*, 1977). Relapse rates averaging about 0.5 per patient-year for the first 5 years are widely quoted (see McAlpine, 1972) although the value of this traditional index of activity has been questioned by Kurtzke (1970) because of the artificial selection of patients with bouts at the expense of those with chronic progressive disease. The 5-year rule (Kurtzke *et al.*, 1977) is a useful prognostic guide. Disability judged on the DSS 5 years after onset correlates (correlation coefficient 0.7) closely with subsequent disability at 10 and 15 years. The 5-year pyramidal and cerebellar scores on the Kurtzke scales were almost as good for predicting 10- and 15-year disability status as the DSS. Furthermore, up to 80% of patients without pyramidal and cerebellar signs at 5 years had DSS scores of only 0–2 at the end of 10 and 15 years

(Kurtzke *et al.*, 1977). Data on long-term survival in MS have been compiled by Kurtzke (Kurtzke *et al.*, 1970) from a large series collected *near outset* and followed for many years. Of 476 patents, all men, who fulfilled the Schumacher criteria for definite MS, the mortality deficit attributable to MS was 8% at 10 years and 20% at 20 years. Age of onset did not influence survival in the 20-year follow-up and only 5% of deaths were unrelated to the neurological disease. Looked at another way, the average life expectancy in young men, after the onset of MS, should be of the order of 35 years, a figure which is in close accord with that obtained more recently by a computerized actuarial approach (Confavreux, Aimard and Devic, 1980). This general conclusion of a relatively favourable outlook of MS in terms of mortality is also borne out by the Danish mortality study of 854 patients in which the average duration of disease was 30 years. In this study (Hyllested, 1961), male mortality was higher than for females, explaining or contributing to the female bias of the sex ratio of cases.

In a review of 342 cases from the Mayo Clinic, remittent types of disease occurred with equal frequency among ambulant and non-ambulant patients (Lazarte, 1950). In this study, progressive and acute disease types were associated with a poorer prognosis, i.e. with the non-ambulant group. A study of 282 patients in Israel showed no difference in prognosis between exacerbating and chronic progressive illness (Leibowitz, Halpern and Alter, 1964). No correlation was found between the duration of initial and second remissions or between age of onset and ultimate disability (Lhermitte *et al.*, 1973). Lhermitte and colleagues calculated that where relapse frequency (0.66 per patient-year) is adopted as a criterion of disease activity, some 100 patients would need to be followed for 4 years to detect an effect of therapy capable of reducing relapses by 25% and this period would need to be extended if chronic progressive disease was also included. Such considerations are clearly relevant for the design of therapeutic trials in MS and demonstrate the extreme difficulty in validating small benefits of treatment. Although prediction of the course of MS on an individual basis is impossible, as it would be for many chronic illnesses, a practical requirement exists to devise straightforward prognostic guidelines incorporating some of the immense accumulation of information on this vexed question. In the writer's opinion, John Kurtzke and his colleagues have gone a very long way towards putting the problem of prognosis in MS on a good scientific footing. MS is fundamentally a chronic progressive disease with a very wide spectrum of disease activity. In the early stages of the illness, the relationship between disability and disease activity is erratic because of the fortuitous effects of randomly distributed lesions. With passage of time, cumulative effects of lesions dominate the clinical picture which more accurately reflects disease activity. These principles are embodied in the 'Kurtzke 5-year rule' which is a statement of the fact that disability 5 years after diagnosis correlates extremely well with later disability and is a more reliable guide to prognosis than relapse

rates or particular system findings. Benign forms of MS including ON, accounting for about 15% of all cases, are identified as such by the '5-year rule'. The poor outlook in hyperacute forms of MS, less than 10% of all cases, is usually clearly apparent well before 5 years and in the event of survival long-term prognosis can still be usefully predicted from disability status (DSS) at 5-years.

The relationship of optic neuritis (ON) to MS is still unresolved. The frequency with which 'idiopathic' acute ON is followed by MS varies considerably in different series. Furthermore, acute ON is commonly encountered in some parts of the world where MS is rare. Subclinical ON, on the other hand, as demonstrated by evoked potentials and other functional tests, is extremely frequent in MS. Acute ON occurring in young adults in regions where MS is common carries a significant risk for MS which doctors are often called upon to discuss with their patients. An early report from the Mayo Clinical (Taub and Rucker, 1954) based on follow-up of 87 patients for 10–15 years indicated that 32% went on to develop MS. This figure reached 40% for patients aged 20–40 at the time of the initial ON. A later study (Percy, Nobrega and Kurland, 1972) on the resident population in Rochester, Minnesota, suggested that only 17% of cases of ON progressed to MS. In the Oxford series (Bradley and Whitty, 1968), 20% of cases of acute ON developed definite MS after a mean follow-up of 10 years. However, a review of 144 cases of ON seen in Belfast showed that 44% of unilateral ON, 65% of bilateral and 55% of recurrent ON went on to MS (Hutchinson, 1976). The risk of MS, derived from McAlpine (1972), is of the order of 40% after 10 years. Acute ON, appearing as a first symptom of MS, was a favourable prognostic sign (Lazarte, 1950). A major study of the prognosis of ON (Compston *et al.*, 1978), in which 146 patients were followed for up to 23 years after onset, again yielded an overall risk of developing MS of 40%. Factors associated with the risk for MS included presence of HLA antigen BT 101, a winter onset and recurrent ON. The presence of oligoclonal IgG in CSF in ON or an abnormal evoked response from the clinically unaffected eye suggest a greater likelihood of later MS.

The term 'benign' MS was employed by McAlpine (1961) to describe a course of disease in which little or no disability developed after at least 10 years. As a group, these cases with a benign course had a high incidence of ON and a low incidence of pyramidal and cerebellar involvement. Asymptomatic forms of MS, with pathological verification as an incidental autopsy finding, also point to a benign form of disease and it has been suggested that up to 20% of autopsy-proven MS was not clinically suspected (Mackay and Hirano, 1967). In general, it appears that three-quarters of clinically definite or probable MS have a 25-year life expectancy and that two-thirds of these patients remain ambulant (*BMJ* Leader, 1972). Such conclusions, based on long-term studies, indicate a better outlook for MS than is commonly appreciated.

Questions about possible adverse effects on the course of MS of events such as pregnancy, illness, trauma or anaesthesia are often asked and may have medico-legal implications. Although there appears to be general agreement that pregnancy does not affect longer-term prognosis (Millar *et al.*, 1959), a number of studies have shown an increased rate of relapse or of onset bouts in the year comprising the pregnancy and puerperium (Millar, 1961; Leibowitz *et al.*, 1967). In a recent review of 512 women, onset of MS and exacerbation of pre-existent illness occurred more than 3 times as often in the 6 months after birth than during pregnancy although, again, long-term prognosis did not appear to be influenced (Posen *et al.*, 1981). The role of oral contraceptives was also considered in this study and had no deleterious effect on the course of MS. The lack of correlation between reported increases in relapse in relation to pregnancy and subsequent disability supports the view (Kurtzke, 1970) that 'bout counting' is of limited usefulness. There are conflicting views about the role of trauma as a precipitating factor in MS. One study of 250 patients demonstrated a history of trauma within 3 months of the first symptoms of MS in 14%, compared with 5% in a control group (McAlpine and Compston, 1952). These findings suggested that trauma might occasionally contribute to onset or relapse in predisposed persons (McAlpine, 1972). Miller (1964) also suggests that exacerbations of MS may rarely follow injuries anatomically related to relevant parts of the neuraxis. Head injury is occasionally noted as an antecedent of unilateral optic neuritis. Severe head or back injuries were associated with exacerbations in 4 out of 255 patients with MS surveyed for possible effects of illness and trauma (Keschner, 1950). Minor trauma, on the other hand, in the form of surgical operations performed under general anaesthesia, does not appear to have an adverse effect on the course of MS (Ridley and Schapira, 1961). A more recent study of the effects of anaesthesia has confirmed earlier impressions that general anaesthesia has no specific effect on the disease but that local spinal anaesthetics do appear to be a risk and may be less preferable to alternative methods in MS (Bamford *et al.*, 1978). There is no clearly defined cause-and-effect relationship between injury and either onset or relapse of MS. Nevertheless, workers' compensation legislation, for instance, is not as a rule concerned with scientific proof of causal relationships but rather with the probability that a particular injury or event contributed to disablement. Every case of this kind has scrupulously to be considered on its merits with particular reference to the site, severity and temporal relationship of injury to subsequent symptoms of MS. Evidence of exacerbation of established MS by injury is not infrequently admitted.

5.6 DIAGNOSIS

The diagnosis of MS cannot be proved neuropathologically during life but is essentially clinical, aided by results of laboratory investigations. Clinical criteria for diagnosis have already been outlined in 5.3.2 (Table 5.3). Evidence

is needed of multiple lesions of CNS white matter and of a profile of disease activity, indicated by patterns of relapse or progression, consistent with MS and for which no other cause can be found. The degrees of probability of clinical diagnosis (Table 5.2) and the problem of incorporation of laboratory data into diagnostic schemata were referred to in 5.3.2. The significance to be attached to laboratory tests in relation to diagnosis or transfer of a patient from one category to another is discussed by McDonald and Halliday (1977) and by Brown *et al.* (1979). Laboratory investigations applicable to diagnosis and which are generally or reasonably widely available are given in Table 5.5.

Table 5.5 Laboratory investigations in MS

1. Evoked responses	Visual
	Auditory
	Somatosensory
2. Blood	Helper/suppressor ratio
	HLA
3. CSF	Mononuclear pleocytosis
	IgG ratio
	IgG index
	Oligoclonal pattern
	IgG synthesis (empirical formula)
	Myelin basic protein
4. CT	
5. Isotope scan	

Elicitation of conduction parameters in CNS pathways by evoked potential methods often provides convincing evidence of subclinical demyelination and, hence, of multiplicity of lesions. Evoked potentials, therefore, delineate more precisely the extent of anatomical involvement by MS and are a subtle extension of the clinical neurological examination. These conduction changes are not disease-specific (Sedgwick, Chapter 7) and clinical interpretation depends *strictly* on the clinical context. Brain scanning (CT, isotope, NMR) is also a useful means of displaying anatomical lesions in MS and providing evidence of multifocal disease. Examination of CSF reveals abnormalities in up to 90% of the clinically definite population (Tourtellotte, Chapter 10) but no aspect of the CSF profile in MS is, in itself, diagnostic of the disease. Study of immunological indices in CSF and blood (Table 5.5) is probably more important in relation to research into pathogenesis and treatment of MS than clinical diagnosis, particularly with the advent of evoked responses and increasingly refined scanning techniques.

A review of CSF findings in MS including cellular changes, intra-BBB IgG production, oligoclonal reaction and a discussion of the IgG ratio and index is provided elsewhere in the book (Tourtellotte, Chapter 10). The finding of a CSF profile 'indicative of MS' aids diagnosis in early or clinically doubtful

cases while unexpected CSF changes alert the clinician to the possibility of other disease. Determination of HLA status is of interest for record and collation purposes but is unhelpful in relation to individual diagnosis. Similarly, from our own limited experience to date, helper/suppressor T cell ratios in peripheral blood appear to have more potential as parameters of immune function in longitudinal studies than as a diagnostic test. Basic myelin protein (MBP) immunoreactive material rises in CSF in acute exacerbations of MS (see Hallpike, Chapter 11) as well as in other CNS diseases causing white matter damage. Radioimmunoassay of CSF MBP is not a routine investigation in MS but is of value as a biochemical marker of myelin breakdown in therapeutic trials. Similarly, studies of immune complexes, proteinase activity, trypsin inhibitors and 2',3'cAMPase (Hallpike, Chapter 11) are motivated by research considerations rather than clinical diagnosis. Abnormal radionuclide brain scans were obtained in patients with MS who were in relapse, while scans were negative in those whose clinical condition was stable (Gize and Mishkin, 1970). The role of computed tomography (CT) in MS has now been extensively validated (Cala and Mastaglia, 1976; Gyldensted, 1976; Aita *et al.*, 1978; Cala, Mastaglia and Black, 1978; Reisner and Maida, 1980). Contrast enchancing lesions are seen in acute MS but more common appearances are of low density areas, particularly in periventricular white matter, and degrees of generalized brain atrophy related to the duration and severity of disease. The CT is, therefore, a valuable potential means of following the course of the disease and complementary techniques such as xenon inhalation are capable of displaying additional lesions (Radue and Kendall, 1978). Very recent introduction of nuclear magnetic resonance (NMR) imaging promises to provide even better resolution of white matter lesions (Doyle *et al.*, 1981). Use of both evoked potential testing and CT suggests that this non-invasive approach permits a firm conclusion to be drawn about diagnosis in 90% of all categories of MS (Mastaglia *et al.*, 1977). A study of evoked potentials (visual and somatosensory) and of CSF immunoglobulin abnormalities (IgG index) has shown that this investigative combination yields diagnostic information in 100% of 'definite', 95% of 'probable' and 80% of 'possible' MS (Trojaborg, Böttchner and Saxtrup, 1981).

5.7 DIFFERENTIAL DIAGNOSIS

An exhaustive account of differential diagnosis of MS would touch upon much of clinical neurology. It is proposed, therefore, to restrict the present discussion to consideration of three topics, namely progressive focal disease, multifocal disease with atypical features for MS, and certain system degenerations. Problems associated with the correct diagnosis of *focal disease* are illustrated by ON and chronic progressive myelopathy. Although progressive visual impairment may occur in MS (Kahana *et al.*, 1973b)

suspicion is always aroused of compressive pathology. Sphenoidal wing and optic nerve sheath meningiomas give rise to progressive painless unilateral visual loss with optic nerve signs which may mimic ON as seen so often in MS. Chronic progression of the visual deficit, without evidence of more widespread CNS disease, is the stimulus for further investigation and the diagnosis of these tumours is usually established by high resolution CT scanning (Moore, Chapter 6; Sanders and Moseley, 1979). Anterior chiasmal notch compression can be misinterpreted as ON when the temporal field defect in the contralateral eye is inconspicuous and overlooked. Visually-evoked responses can provide evidence of demyelinating disease if there are marked latency delays but must be interpreted with caution as somewhat similar abnormalities also result from compression of the anterior visual pathways (Halliday *et al.*, 1976). The importance of serial visual field testing and repeated neuroradiology in doubtful cases cannot be over-stressed. Although MS is one of the commonest causes of progressive spastic paraparesis of middle life, the diagnosis is by exclusion of other disease unless there are signs of disseminated lesions or a history of past demyelinating episodes. In one series, application of visually-evoked responses, examination for subtle neuro-ophthalmological signs and CSF analysis, enabled three-quarters of the patients presenting with chronic myelopathy to be classfied as MS (Bynke, Olsson and Rosén, 1977). It is of interest that in another group with chronic myelopathy of undetermined cause (Link, Norrby and Olsson, 1976) a majority of patients had oligoclonal IgG in CSF but that there was no difference in the course of the disease between those who did and did not have the oligoclonal change. Presence of pain and finding of a trunk sensory level are indications for myelography. Paraparesis of subacute onset occurs with extramedullary and intrinsic cord tumours, with some thoracic intervertebral disc protrusions (Carson, Gumpert and Jefferson, 1971), arachnoiditis and angiomas. Spinal vascular malformations deserve particular mention because episodic progression of disability with these lesions as well as exacerbation by trauma or pregnancy may lead to erroneous diagnosis of MS (Aminoff and Logue, 1974). Myelopathy with cervical spondylosis, associated with narrowing of the spinal canal (Nurick, 1972) or bulging of the ligamentum flavum (Stoltmann and Blackwood, 1964), may be difficult to distinguish from MS and the conditions may clearly also co-exist.

Multifocal disease of the nervous system which is not MS falls into two categories: (1) illness with multisystem involvement and (2) illness confined to the CNS. In the multisystem group, connective tissue disorders may cause nervous system involvement resembling MS (Fulford *et al.*, 1972). Systemic lupus erythematosus (SLE), running a chronic relapsing course, occasionally gives rise to transient cerebral phenomena, dementia or myelopathy due to immune complex vasculitis. A picture of multifocal CNS involvement may also be given by sarcoidosis. Behcet's syndrome, a condition of unknown aetiology with prominent muco-cutaneous and ocular involvement, may

present a varied neurological picture with a confusional state, meningomyelitic syndrome, brainstem signs and a marked polymorph pleocytosis in CSF (Pallis and Fudge, 1956; Mason and Barnes, 1969). In the second group, confined to the CNS, so-called granulomatous angiitis is an extremely rare but seemingly distinctive entity. The correct diagnosis of granulomatous angiitis has only infrequently been made during life but the fluctuating downhill course with multiple CNS deficits arouses suspicion of an acute type of MS (Harrison, 1976). The CSF findings in this disease have been reviewed by Fishman (1980) and are by no means always clearly distinguishable from those found in MS. A case of relapsing myelopathy due to this unusual form of vasculitis has now been reported (Rawlinson and Braun, 1981). 'Central neurofibromatosis' is a rare entity in which multicentric tumour development may initially give rise to syndromes capable of being confused with MS.

System degenerations capable of giving rise to confusion with MS are mainly in the group of inherited ataxias. When these conditions manifest in dominant pedigrees as for instance in Marie Tooth disease, hereditary spastic paraplegia, elicitation of the family history will point clearly towards the correct diagnosis. The recessive diseases pose greater problems unless the overall clinical features, as for instance in Friedreich's ataxia, are plainly disparate from MS. Pes cavus is a useful clinical clue to the presence of the inherited disorders. Leber's hereditary optic atrophy is mentioned elsewhere (Moore, Chapter 6). Abnormal visually-evoked responses are frequently found in the hereditary ataxias and this form of investigation cannot be readily employed to distinguish these diseases from MS.

5.8 CONCLUSIONS

The clinical features of MS are an essential framework for new initiatives in the arenas of therapy and management as well as for research into causation and pathogenesis. The variable manifestations and course of MS are seen as expressions both of the effects of individual lesions as well as of disease activity or 'turning-on' and 'turning-off' of the pathological process. For clarity of presentation, an attempt has been made to represent MS as a chronic, immunologically-mediated, inflammatory disease of the CNS in which fluctuating symptomatology reflects siting of demyelination and where longer-term disablement is a function of the intrinsic severity of the disease process in the individual patient. Support for such a concept is provided by evoked potential studies in MS which indicate a remarkably high degree of subclinical involvement of the CNS. Evidence of persistent immunological abnormalities in MS centre on the CSF changes and, in particular, the frequency of oligoclonal IgG which clearly mirrors some aspect of immune pathology. In spite of the distinctive clinical features of MS, failure to identify an infective or metabolic cause points increasingly towards the central importance of immune dysregulation and to the possibility of a syndrome

Evoked potential studies

based on interplay between relatively trivially provocative factors and a 'susceptible' immune apparatus. The increasing sensitivity of immunological probes and the recent finding of altered T cell subset ratios in MS are promising pointers for the future.

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CHAPTER SIX

Neuro-Ophthalmological Aspects of Multiple Sclerosis

Colin E. Moore

6.1 Optic neuritis
6.2 Chiasmal and retro-chiasmal lesions
6.3 Periphlebitis retinae and peripheral uveitis
6.4 Pupillary signs
6.5 Eye movement abnormalities

Patients who develop multiple sclerosis (MS) often present initially to the ophthalmologist with impaired vision due to retrobulbar or optic neuritis (ON). Less frequently, presentation is with double vision due to extraocular muscle (EOM) palsy. The most commonly encountered physical signs are those of active or sublinical ON, nystagmus and internuclear ophthalmoplegia (INO).

6.1 OPTIC NEURITIS

The usual presenting complaint is of unilateral visual impairment for a few days or a week associated with discomfort in or around the affected eye which is worse on eye movement. Pressure over the superior rectus insertion may cause pain because of traction on the inflamed posterior optic nerve. The visual change may be described as mistiness, blurring, fogging or decreased brightness. Usually, vision is not lost completely. Deterioration to light perception only is quite rare, as is simultaneous involvement of both eyes. Consecutive involvement of each eye in turn occurs relatively frequently. Depth perception with ON is impaired, causing difficulty judging steps and a moving ball in games such as squash, tennis or cricket. Positive visual phenomena (phosphenes) produced by eye movements, and recognized in normal people and myopes with vitreous opacities, have also been observed in patients with ON and are thought to be analogous to the parasthesiae of the trunk or extremities occurring with quick neck flexion (Lhermitte's phenomenon) in patients with multiple sclerosis and other diseases of the posterior columns (Davis *et al.*, 1976). Although pupil size is usually unaffected in acute ON, an afferent pupillary defect (best displayed by the swinging flashlight test) is constantly seen. A thin slit lamp beam of light focused at the pupillary margin will induce regular persistent oscillations of

has been extensively studied and varies in published series between 15% and 85%, depending on the methods of selection and the incidence of MS in the community from which the patients are drawn. The probability is that a lower rather than higher percentage develop MS (Bradley and Whitty, 1968). There is also some evidence that *acute* ON may be associated with more benign patterns of MS (Percy, Nobrega and Kurland, 1972). Cohen, Lessell and Wolf (1979) prospectively studied 60 patients with uncomplicated ON for at least five years to determine the subsequent risk of MS. Seventeen of these patients (28%) developed definite MS and four (7%) developed probable or possible MS. Six of the seventeen who developed definite MS did so within the first year and 45% of the women, but only 11% of the men, developed MS. Both sexes were at highest risk if the ON developed between 21 and 40 years with 51% of patients in this age group progressing to MS, whereas the risk for the others was 12%. There was an over-all increased risk of MS with recurrent ON. If 'slits' are seen in the nerve fibre layer in the fundi of patients presenting with acute ON, the subsequent development of MS is much more likely.

In another series, 40% of patients reassessed 1 month to 23 years after presentation with isolated optic neuritis went on to develop MS (Compston *et al.*, 1978). It was suggested that ON is caused by two environmental agents or groups of agents and that the agent which is most common in the winter leads to the development of MS in the genetically susceptible individual. The agent more common in the summer is much less likely to cause MS in either susceptible or non-susceptible individuals. The presence of HLA antigens associated with MS confers a specific disadvantage on individuals in their ability to handle infection by the putative MS causative agent thus permitting damaging immunological processes to develop. In an attempt better to define the likelihood of developing MS after ON, a nationwide study of ON was carried out in Israel (Kahana, Alter and Feldman, 1976). As with MS, ON was more common in European immigrants than in Afro-Asian immigrants. During a follow-up interval which ranged from 3.3 to 15.6 years, 28% of 105 patients developed MS. This study supported earlier work using life-table methods carried out in Hawaii, which showed that between 29 and 39% of patients with ON will develop MS within 10 years of onset.

Management of acute ON due to demyelination differs between clinicians and ranges from no treatment at all (Hoyt; personal communication) to active treatment with oral corticosteroids, or ACTH given intra-muscularly (Rawson, Liversedge and Goldfarb, 1966; Rawson and Liversedge, 1969). Unilateral ON is best not treated (Lessell, 1974). Patients may recover quickly from ON when treated with corticosteroids or corticotrophin but over the longer term (one year) no convincing differences exist in degree of visual recovery between treated and untreated patients (Bowden *et al.*, 1974). Nevertheless, it seems reasonable to give corticotrophin or prednisolone when there is considerable pain or incapacitating visual loss. The injection of

steroid into Tenon's capsule and behind the globe in the retro-bulbar space has been advocated (Gould *et al.*, 1977). Such procedures are not generally recommended, however, because of the lack of proof of any long-term benefit (Bird, 1976), and because of a significant risk even in skilled hands, of causing perforation of the globe or optic nerve damage.

6.2 CHIASMAL AND RETRO-CHIASMAL LESIONS

Demyelination affecting the optic chiasm must be remembered as one of the rare non-surgical causes of a bitemporal hemianopia (Schatz and Schezinger, 1976). Spector, Glaser and Schatz (1980) described six patients with demyelinating chiasmal lesions. They found a predilection for females in the third to fifth decades, modest to marked visual field defects of a chiasmal pattern, and with generally good recovery. Demyelination affecting the tract and lateral geniculate body (LGB) is possible in MS. Tract lesions are characterized by definitely incongruous hemianopic field defects while LGB lesions produce smaller incongruous hemianopic wedge-shaped defects, or characteristic incongruous defects of irregular geographical outline when the hemianopia is incomplete. While an instance of demyelination definitely involving the LGB has not yet been described in the literature, LGB involvement by tumour, arterio-venous malformation, necrosis and vascular infarction have been annotated.

In an excellent review paper, Hawkins and Behrens (1975) report bilateral homonymous hemianopia in a patient with MS. These authors cite Uhthoff, who commented in 1890 on the frequency of pathological lesions in the posterior visual pathways without the usual clinical manifestations of MS. Savitsky and Rangell (1950) found asymptomatic lesions in the optic radiations in 23 of 50 autopsies but no homonymous field defects in 415 personally observed patients. Hawkins and Behrens (1975) offer various reasons for this disparity between pathological and clinical manifestations, namely, small lesions in widespread fibres in the radiations, physiological differences between pregeniculate and retrogeniculate pathways, involvement of the more peripheral uniocular visual field fibres close to the ventricles, as well as lack of critical testing in otherwise severely handicapped patients.

Abnormal isotope brain scans are rarely seen in MS. A positive scan is seen only with acute localized retrogeniculate demyelination (Moore, 1977; Gize and Mishkin, 1970; Moses, Davis and Wagner, 1972; Cohen, Fermaglich and Auth, 1975; Antunes, Schesinger and Michelson 1974; Miller and Potsaid, 1974). Positive brain scans have been reported in Schilder's disease (Valenstein, Rosman and Carter, 1971) and progressive multifocal leuco-encephalopathy (Mosher, Schall and Wilson, 1971). More MS plaques are revealed by CT scanning than by radionuclide imaging. Some 30% of patients with moderate or severe symptoms of MS have plaques demonstrable on CT scanning (Cala and Mastalgia, 1976; Weinstein *et al.*, 1978). The findings

consist of low attenuation lesions situated mainly in the white matter which do not produce space-occupying effects, ventricular dilation with irregular edges, and enlarged sulci. Contrast enhancement is thought to be caused by changes in the blood–brain barrier, perhaps occurring transiently during the evolution of such lesions. Repeated scanning and angiography enable differentiation from tumours. Presence of enhancing lesions at sites unrelated to clinical features provide evidence of multiple central nervous system pathology (Harding, Radne and Whiteley, 1978).

6.3 PERIPHLEBITIS RETINAE AND PERIPHERAL UVEITIS

In 1944, Rucker described white peri-venous sheathing in 34 patients, 21 of whom were diagnosed as having definite MS, and in seven more who were suspected of having the disease. The condition also affected the peripheral veins and sometimes a plaque was seen overlying the vessels. There was no history of previous optic neuritis and arteries were unaffected. The sheathing is seen only with difficulty, utilizing fully dilated pupils and an indirect ophthalmoscope light (Rucker, 1972; Arné *et al.*, 1978). This periphlebitis gives support to the long standing suggestion that plaques in MS have their origin around veins in the central nervous system (Putnam and Adler, 1937). Peripheral uveitis, characterized by vitreous opacities and inflammation of the pars plana of the ciliary body, is said to occur in 15%–27% of patients with MS, possibly antedating MS by many years (Archambeau, Hollenhorst and Rucker, 1965; Breger and Leopold 1972; Brockhurst, Schepens and Okamura, 1960; Giles, 1970; Curless and Bray, 1972; Porter, 1972).

6.4 PUPILLARY SIGNS

These are not characteristic, apart from the relative afferent pupillary defect in RBN. Walsh and Hoyt state that there is difference of opinion as to whether true Argyll Robertson pupillary reactions occur in MS but partial Argyll Robertson pupils are certainly possible. A dilated pupil with a nuclear third nerve lesion is quite rare, as is Horner's syndrome.

6.5 EYE MOVEMENT ABNORMALITIES

Nystagmus is one of the most common physical signs in MS. It is often present early in the disorder and tends to be permanent. The nystagmus can be of various types. Acquired pendular nystagmus in adults, suggesting cerebellar nuclear lesions (Daroff *et al.*, 1978), is usually of dissociated type in MS. Acquired horizontal jerk nystagmus is thought to be due to involvement of the vestibular nuclear complex in the brainstem. Ataxic nystagmus indicative of INO is discussed below. Circular elliptic nystagmus may be seen in MS and is often dissociated in the two eyes and almost always co-existent with truncal or

extremity ataxia (Daroff *et al.*, 1978). Various cerebellar system eye signs may be present, including cogwheel (saccadic) pursuit movements, hypometric saccades, slow saccades, skew deviation, asthenia of upward gaze, square wave jerks and macro-square wave jerks, macro-saccadic oscillations, upbeat, downbeat, positional, rebound nystagmus, ocular dysmetria, ocular flutter, opsoclonus and ocular myoclonus. Vertical gaze palsies are sometimes observed. An instance has recently been described (Slyman and Kline, 1981) of the occurrence of a dorsal midbrain syndrome in MS, i.e. upward gaze paresis, convergence retraction nystagmus, light-near dissociation and skew deviation.

Internuclear ophthalmoplegia (INO) is an important eye movement abnormality which is generally due to MS when it is present bilaterally. Impulses originating in the pontine paramedian reticular formation (PPRF), the horizontal 'gaze centre', pass to the ipsilateral VI nucleus, and fibres which cross to the other side in the pons ascend in the medial longitudinal fasiculus (MLF) to the medial rectus subnucleus in the midbrain (see Fig. 6.2). A lesion which interrupts these ascending fibres in the MLF to one side of the midline produces unilateral impairment of adduction (on the same side) and a characteristic horizontal nystagmus in the abducting eye, the so-called 'ataxic nystagmus'. Convergence is preserved. The reason for ataxic nystagmus is not known with certainty. Impaired inhibition of the medial rectus of the abducting eye has been suggested as a cause, or convergence acting intermittently on the medial rectus of the abducting eye (Pola and Robinson, 1976; Gay *et al.*, 1974). Adaptive pulse step mismatching (Baloh *et al.*, 1978) may also explain the occurrence of dissociated or ataxic nystagmus in MLF lesions.

Pseudo-INO is seen in myasthenia gravis, without ataxic nystagmus. The most frequent cause of INO in young adults, particularly when the disorder is bilateral, is MS. However, I have seen unilateral INO become bilateral over 24 hours in a 19-year-old female with disseminated lupus erythematosus. In older persons occlusive vascular disease may produce INO, which is then usually unilateral. Other rare causes are diabetes, encephalitis, intrinsic tumours, e.g. glioma, Wernicke's encephalopathy and syringobulbia. Subtle INO may be brought out with horizontally moving optokinetic targets or by testing for ocular dysmetria, in which there is undershoot of the adducting eye and overshoot of the abducting eye. Abnormality of the saccadic horizontal movements should also be looked for. Glaser (1978) says that classifying INO into anterior or posterior types is of no practical value to the clinician. Rare cases of the posterior type of INO due to lesions between the centres for conjugate gaze and the abducens nuclei are categorized as posterior INO of abduction. A case is reported in which there is slowing of abduction saccadic movement without limitation of end movement (Kommerell, 1975) so the saccadic and smooth pursuit eye movements as well as end positions should be carefully observed.

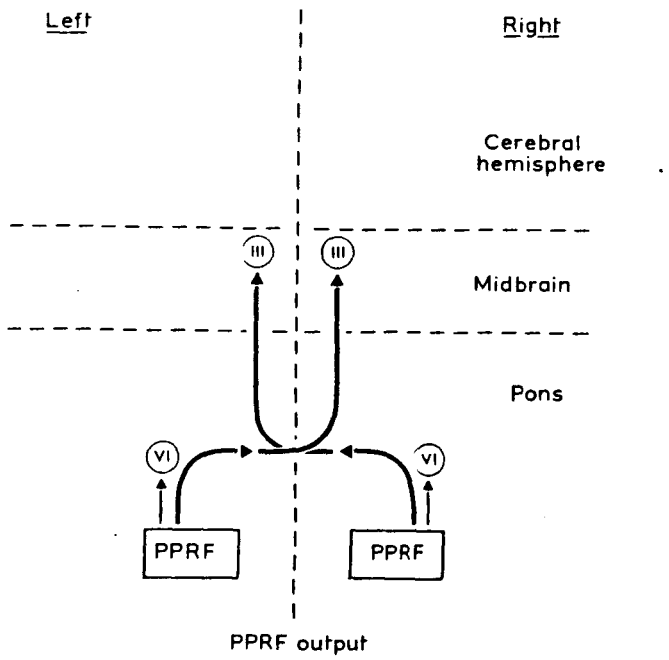

Fig. 6.2 Reprinted with permission from Duane's *Clinical Ophthalmology* vol. 2, Chapter 10, p. 2, Fig. 10.4.

A lesion of the PPRF and the ipsilateral MLF produces a horizontal gaze palsy to the affected side and INO on gaze to the opposite side, the '*one and a half*' syndrome. This is most commonly seen as a result of vascular disease of the brainstem but may also occur in demyelination. External ocular muscle palsies are fairly frequently seen in MS, with diplopia as the presenting symptom. One or several muscles may be involved. Diplopia often occurs before a frank limitation of movement is apparent. Ptosis, partial oculomotor paresis, sixth nerve palsy, and other pareses have been described (Kahana, Leibowitz and Alter, 1973). The fourth nerve is rarely, if ever, affected. However, histological changes have been found in the fourth nerve nucleus in MS.

The ocular manifestations in MS have been briefly surveyed. The neuro-ophthalmologist, besides helping to manage ocular symptoms of MS, can aid in the diagnosis and understanding of the disorder by detecting optic nerve head changes, nerve fibre bundle and related visual field defects, peri-venous exudates, uveitis, characteristic and sometimes subtle eye movement abnormalities (Solingen *et al.*, 1977), and by analysing more elusive symptoms and signs attributable to chiasmal and retrochiasmal demyelination.

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CHAPTER EIGHT

The General Pathology of Multiple Sclerosis: Morphological and Chemical Aspects of the Lesions

C. W. M. Adams

8.1 General nature of the lesion
8.2 The typical lesion
8.3 The Marchi osmium reaction
8.4 The active lesion
8.5 Early lesions
8.6 Variants of the plaque in multiple sclerosis
8.7 Diffuse lesions in multiple sclerosis
8.8 The continuous pathological process in multiple sclerosis
8.9 Correlation of plaques with clinical history
8.10 Neuroglia in multiple sclerosis
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8.13 Changes in brain lipids
8.14 Proteins, lipoproteins and proteolytic enzymes
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It is not proposed here to dwell upon the historical aspects of multiple sclerosis and the early reports in the 19th century. Rather it is intended to concentrate on certain pathological features that seem to be relevant to the initial changes and spread of the process. The pathological appearances of multiple sclerosis have been widely described in a number of careful and extensive studies, often based on large numbers of cases (e.g. Dawson, 1916a, b; Adams and Kubik, 1952; Adams and Richardson, 1961; Jellinger, 1969 (85 cases); Lumsden, 1970 (83 cases); Guseo and Jellinger, 1975 (143 cases)). It is proposed here to exploit much of the excellent material in these studies, as well as the 107 cases now collected in the Multiple Sclerosis Tissue Bank supported by the Medical Research Council at Guy's Hospital Medical School, London. Part of this last series has previously been reported (Adams 1975, 1977).

As mentioned above, the main aim of this chapter is to present those histopathological features which are relevant to the pathogenesis of the lesions, to consider the extension and enlargement of lesions, and, if possible, to adduce some information about the aetiology of the disease.

8.1 GENERAL NATURE OF THE LESION

It is widely recognized that the distinctive feature of a primary demyelinating disease is destruction of the myelin sheath, but with preservation of the axonal process. Charcot (1868, 1877) was the first clearly to show this in multiple sclerosis, and his observation was convincingly supported by the subsequent biochemical finding of persistent axonal acetylcholinesterase activity in the lesion (Lumsden, 1965). Histochemical evidence shows some acetylcholinesterase activity in the lesions (non-specific cholinesterase is mainly in the neuroglial cells), but it must be admitted that the results are not really adequate for comparative studies.

Swellings, torpedoes and more advanced evidence of axonal degeneration are sometimes seen in chronic lesions but, in general, axons are initially intact and considerably spared even quite late in the development of the plaque. This is, of course, consistent with the clinical observation that signs and symptoms usually largely remit at 6–12 weeks after the onset of a particular episode and that much of the functional defect is due to inflammatory oedema around the lesion. However, persistent clinical features must be held to result from impaired saltatory conduction caused by loss of myelin or failure to replace an adequate amount of it (see Harrison, Chapter 14).

8.2 THE TYPICAL LESION

The characteristic plaque of chronic multiple sclerosis appears on gross examination of the brain and spinal cord as a sharply defined, greyish-yellow, rubbery, semi-translucent, circular or elongated lesion.

A yellow colour may be imparted to the plaque if much lipofuscin is present (Adams, 1975). More recent plaques are less rubbery and may be rather grey and slightly pink in colour, and are less distinct in outline (see below). Plaques commonly involve the central white matter but, according to Lumsden (1970), plaques in the cortex and subpial regions of the gyri numerically outnumber those in white matter by four to one. Periventricular plaques (involving the lateral and fourth ventricles) are exceedingly common and are found in over 90% of cases: such lesions were found in all but five of the last 54 cases in our tissue bank. Plaques are also commonly found in the optic tracts and frequently in the cord, pons, mid brain and basal ganglia. Lesions are in general not symmetrical, but a number of occasions were recorded by Lumsden (1970) where symmetrical plaques were found in the cord and pons. Plaques are about twice as common in the cervical than in the lower part of

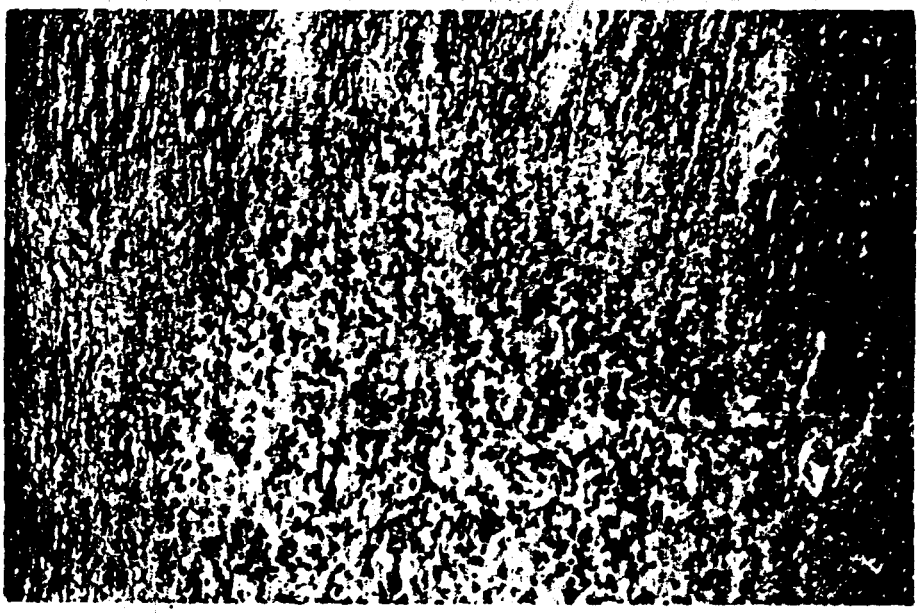

Fig. 8.1 Acute plaque of multiple sclerosis showing lipid-containing mononuclear phagocytes throughout the lesion. Note the shelving edge. Sudan black. ×43.

the cord. This difference has been ascribed to stress engendered by the denticulate ligaments inserted into the lateral surfaces of the cervical cord (Oppenheimer, 1978).

The frequent involvement of the periventricular region calls for special comment: lesions in this site appear to be particularly old in their appearance, as though they are part of the original pathogenic process. The lesion wraps around part of the circumference—usually one-third to one-half—of the ventricle; the lateral and fourth ventricles are more commonly involved than the main body of the third ventricle.

It has long been realized that the characteristic plaque of multiple sclerosis encircles a venule or small vein, from which it appears to have arisen (see below). This relation to small veins has been convincingly confirmed by serial section studies, where plaques were examined in their continuity and their relationship to veins explored (Fog, 1964, 1965). Often plaques extend for a considerable distance along the course of a vein; a wormlike plaque is particularly characteristic of the spinal cord (Lumsden, 1970).

Histologically, the common chronic plaque of multiple sclerosis shows a clear-cut edge in myelin preparations (e.g. Luxol fast blue) but, as mentioned above, many axons passing through the lesion are seen to persist with silver impregnation methods. If the lesion is still active, some foam cells (gitterzellen) may be seen at its edge, together with sudanophilic Marchi-positive breakdown products of myelin (Figs 8.1 and 8.2). Activity is also indicated by the presence of a glial reaction, which encompasses astrocytic

Fig. 8.16 Perivenular lymphocytoid infiltration in active multiple sclerosis. Note Russell body (arrow) and plasma cells. Haematoxylin and eosin. ×595.

whether the lymphocytic reaction does not start in some cases in the meninges and travel retrogradely back to the brain parenchyma along the Virchow-Robin space around small veins and venules. Perhaps Lumsden (1970) was thinking of a similar possibility when he described the infiltration of immunoaggressive lymphoid cells from the meninges into the Virchow-Robin space. Their travel in this perivascular space would be facilitated by the lymphatic channels which have been further characterized there (Prineas, 1979).

Apart from the moot question of the nature of the specific oligoclonal antibodies secreted by lymphocytes and plasma cells within the central nervous system in multiple sclerosis, these perivenular infiltrates have provoked much interest because of their possible implication in viral or autoimmune disease. Could it be that a modest acute perivenular viral encephalomyelitis may either resolve or become chronic, perhaps because of a superimposed autoimmune reaction? In this connection, the resemblance has often been noted between certain stages of active multiple sclerosis, experimental allergic encephalomyelitis, perivenular viral encephalomyelitis (Adams and Richardson, 1961; Adams and Sidman, 1968) and post-rabies-vaccinial encephalomyelitis (Uchimura and Shiraki, 1957). However, as observed by Leibowitz (Chapter 12), histological appearances may suggest but never prove relationships.

Fig. 8.17 Surface of lateral ventricle with early periventricular lesions of active multiple sclerosis. Note sleeves of demyelination following the course of parallel rows of subependymal venules. V = venules with surrounding pale areas of demyelination; E = ependymal surface; C = cut surface. Stained in the gross with nile blue sulphate, ×3.

8.12 PERMEABILITY AND RELATION OF LESIONS TO VENTRICLES AND VEINS

As already mentioned an old chronic periventricular lesion is a frequent—if not universal—feature of multiple sclerosis: it is as though this lesion was a component of the original pathogenic mechanism (see Fig. 8.17). The frequency of periventricular lesions and the ubiquitous perivenular lesion strongly suggest that some substance, agent or cell may originally have penetrated into the brain from the cerebrospinal fluid (through the meninges or Virchow-Robin space) or from venous blood or through both routes (Adams, 1972). Antibody formed by immunologically competent cells in the recesses of sulci (see above; Fig. 8.18; Adams, 1977) might more easily penetrate the brain from the ventricular system, particularly in the vicinity of the choroid plexus. Antibody in the blood or some other agent might be expected more readily to enter the brain if vascular permeability is increased.

Fig. 8.18 Perivenular infiltrate and lymphocytic meningitis involving a sulcus in active multiple sclerosis. Note numerous plasma cells. Haematoxylin and eosin. ×119.

The endothelium is the main structure concerned in the control of vascular permeability. Such control is exerted by the tight junctions between endothelial cells and, for larger molecules, by the rate of vascular transport. Permeability is increased in the brain in experimental allergic encephalomyelitis (Leibowitz and Kennedy, 1972), and fenestrations have been described in cerebral capillary endothelium in the chronic form of the disease (Snyder, Hirano and Raine, 1975). These observations suggest that permeability might be increased around the perivenular lesions in multiple sclerosis, and, certainly marked oedema is sometimes seen around these perivenular infiltrates (see Figs 8.6 and 8.7). Moreover, Broman (1947, 1964) demonstrated increased permeability of vein walls in the plaque to dyes such as Evans blue, even though the plaque vessels show normal histoenzymic staining with alkaline phosphatase and non-specific adenosine triphosphatase (Adams, Hallpike and High, unpublished 1970). Tourtellotte and Parker (1967) found no increased permeability in the plaque to albumin, Br^-, Cl^-, Na^+ and K^+ However, it is not recorded whether these results applied to lesions with perivenular infiltrates, for these would be those that might be expected to show an increase in permeability.

Mast cells have been identified in the perivascular regions of the brain (Ibrahim, 1974). Such cells are a potent source of vasoactive peptides, and

such compounds are obviously relevant to permeability changes in what is in effect a perivenular vasculitis in multiple sclerosis.

Hyalinized small vessels are frequently seen in chronic plaques of multiple sclerosis as well, occasionally, outside the defined lesion. The nature of these vessels is not altogether clear, as to whether they are thick-walled sclerotic veins or small arteries without an obvious internal elastic membrane or both sorts of vessel. Similar findings have been recorded by Allen (1981). The effect of such hyalinization on permeability is not known, but it is well-recognized, for example, that hyalinization of small vessels in diabetes mellitus actually leads to increased permeability (see Adams, 1967). The presence of such hyaline material could be taken as evidence of a previous or mainly burnt-out vasculitis in that region.

8.13 CHANGES IN BRAIN LIPIDS

For many years the cause of multiple sclerosis was sought in some change in the myelin lipids. It had been considered that myelin was metabolically unstable, then it was shown that cholesterol turnover was exceedingly sluggish in myelin (Davison *et al.*, 1959). Opinion then swung away from metabolic instability and centred on some specific change in the myelin lipids. However, apart from changes in linoleic acid and secondary chemical degradation, no convincing primary lipid alterations have been shown in the myelin in multiple sclerosis. Lipid deposits of metachromatic lipid have been noted in apparently normal myelin outside plaques (Bayliss and Adams, 1971), but their origin remains unexplained. Other lipid abnormalities were at first partly attributed to Wallerian degeneration, but it is more probable that they represent small foci of multiple sclerosis. 'Normal white matter' is a very dubious description in multiple sclerosis, as small barely visible lesions are considerably more common than the typical established plaque (see above), and esterified cholesterol is frequently present in the apparently normal white matter in multiple sclerosis (Wender *et al.*, 1973; see Norton, 1977).

The formation of esterified cholesterol from unesterified or free cholesterol in the myelin sheath has been regarded as the hallmark of demyelination (Cumings, 1953, 1955). This is undoubtedly true, but it is a secondary event (Adams, Ibrahim and Leibowitz, 1965) which is a function of the mononuclear phagocytes in multiple sclerosis (Petrescu, 1969). These cells are equipped with the enzyme acylCoA: cholesterol acyl transferase to carry out such esterification (see Day, 1964). However, the degradation of myelin is a relatively slow process in the central nervous system (McCarman and Robins, 1959; Lampert and Cressman, 1966; Bignami and Rolston, 1969; see Lampert, Chapter 2). Thus, the formation of esterified cholesterol is a delayed event that has no direct relevance to initial mechanisms in demyelination. Whether or not the ingestion of myelin fragments by mononuclear phagocytes is a primary event in multiple sclerosis remains to be elucidated and is

discussed above (Section 8.10 on Neuroglia in multiple sclerosis; see also Lampert, Chapter 2 and Raine, Chapter 13).

Intermittent interest has been taken over the last 30 years in the possible role of polyunsaturated fatty acids in multiple sclerosis (Swank *et al.*, 1952). It was later shown that subjects with multiple sclerosis have a deficiency in linoleic acid in esterified cholesterol in their serum and brain (Baker *et al.*, 1965, 1966). Red cell lipids are also abnormal in that the usual inverse ratio between linoleate and arachidonate is not maintained. For these reasons, efforts have been directed towards treating multiple sclerosis with supplements of polyunsaturated fatty acids. However, the clinical results (Millar *et al.*, 1975) have shown only a marginal benefit, with improvement in the severity of relapses but with no long-term improvement in prognosis (see Hallpike, Chapter 11).

On another tack it has been shown that linoleic acid is an immunosuppressant. Any therapeutic success with linoleic acid might, therefore, depend upon the damping-down of some ill-directed immune process rather than the correction of a fatty acid deficiency. The increased platelet stickiness in multiple sclerosis (Caspary *et al.*, 1965; Wright, Thompson and Zilkha, 1965; see Hallpike, Chapter 11) might also be related through linoleic acid to some change in a platelet-active prostaglandin (e.g. thromboxane), but this remains a very speculative area. The increased platelet stickiness is relevant to Putnam's 1937) proposed thrombogenic aetiology of multiple sclerosis. However, the occurrence of thrombosis within the plaque veins has not been confirmed and was probably only post-mortem blood-clot. Moreover, there is no increased incidence of deep-vein thrombosis in multiple sclerosis patients, even in spite of their greater risk during immobility (data from the present series of 106 cases). Likewise, multiple sclerosis patients have no particular risk for ischaemic heart disease or cerebral infarction (present data; and Allen, Millar and Hutchinson, 1978).

The likelihood of a disorder in myelin lipids as the *primary* fault in multiple sclerosis can be seen from the foregoing discussion to be fast receding. This represents a remarkable volte-face in attitudes among research workers over the last 20 years.

Reports on the recovery of intact prepared myelin from multiple sclerosis plaques and, in particular, the essential lack of any abnormality therein cannot mean that the myelin in such plaques is normal. For example, Cuzner *et al.* (1976 pointed out that myelin basic protein is deficient in the plaque, as shown by polyacrylamide-gel electrophoresis, but prepared myelin shows no such abnormality. It is likely that the initial effect of the demyelination process is to cause physical fragmentation of myelin, as is seen in the preliminary phase of Wallerian degeneration (Johnson *et al.*, 1950; Adams, 1962a). Such physical fragmentation is probably caused by disintegration of the protein skeleton of the myelin lamella (Adams and Tuqan, 1961a; Adams, 1962a, b; Hallpike and Adams, 1969). It is not unreasonable to propose that profound

changes in the physical structure of myelin would alter its behaviour o density-gradient preparations. Perhaps partly fragmented myelin no longer separates at the same density as does normal myelin. Hence, only normal myelin would be recovered from the plaque, and abnormal myelin would disintegrate or be recovered at another density.

8.14 PROTEINS, LIPOPROTEINS AND PROTEOLYTIC ENZYMES

It has been known for some years that the activity of proteolytic enzymes increases during Wallerian degeneration (Porcellati and Curti, 1960; Adams and Tuqan, 1961a; Adams, 1962a, b; Hallpike, Adams and Bayliss, 1970a). It was observed that a proteolytic enzyme can induce '*in vitro demyelination*' by removing the protein skeleton from myelin in frozen histological sections of white matter (Fig. 8.19; Adams and Tuqan, 1961a). A histoenzymic study, using the gelatin autogram method, showed that proteolytic enzyme activity increases in and around plaques of multiple sclerosis (Adams, 1968). Likewise, plaques show increased fibrinolytic activity with Todd's fibrin autogram method (Hirsch, Blanco and Parks, 1981). Biochemical and histochemical studies revealed that basic protein is removed from plaques and that cerebral acid proteinase increases, not only in the lesion, but also in the surrounding white matter (Hallpike and Adams, 1969; Einstein *et al.*, 1970, 1972; Hallpike, Adams and Bayliss, 1970b, c; Riekkinen *et al.*, 1970a, b; 1971; Adams *et al.*, 1971; Hirsch *et al.*, 1976; Roytta *et al.*, 1976; Newcombe *et al.*, 1980; and others). Partial degradation of myelin basic protein by such proteolytic enzyme may allow a soluble smaller peptide, carrying the antigenic determinants of the larger molecule, to reach immunoreceptor sites outside the nervous system (Einstein, Csejtey and Marks, 1968). In this way an

Fig. 8.19 Tryptic digestion removal of myelin lipids from frozen section of human brain. Left, Sudan black; right, toluidine blue (non-specific stain). ×3. In part reproduced with permission from *J. Neurochem.*, 1961, 6, 327).

autoimmune reaction could be started and perpetuated. However, circulating antibodies against myelin basic protein have not so far been detected in patients with multiple sclerosis (but see Leibowitz. Chapter 12). Nevertheless, basic protein does seem to be a particularly vulnerable constitutent of myelin, in that it appears to be a sort of structural glue that holds the consituent lamellae together and that it is particularly susceptible to proteolytic enzymes and autolysis (Einstein *et al.*, 1968; Marks, Benuck and Hashim, 1974; Banik, 1979). Thus, the breakdown of myelin in both multiple sclerosis and Wallerian degeneration might be mediated by proteolytic enzymes, directed against the protein skeleton of the myelin sheath, as diagramatically represented previously (Fig. 8.20; Adams, 1962b . Two arguments could be raised against this conclusion. First, that some other protein apart from the basic sort may be involved. For example, a myelin-associated glycoprotein has been shown by immunoperoxidase histochemistry to be lost from a wider area around the multiple sclerosis plaque than is basic protein and stainable lipid (Itoyama *et al.*, 1980). In addition changes in myelin protein in multiple sclerosis, apart from basic protein, have been described by Csejtey *et al.* (1975) and by Newcombe *et al.* (1980). Second, it is not certain that a deficiency of

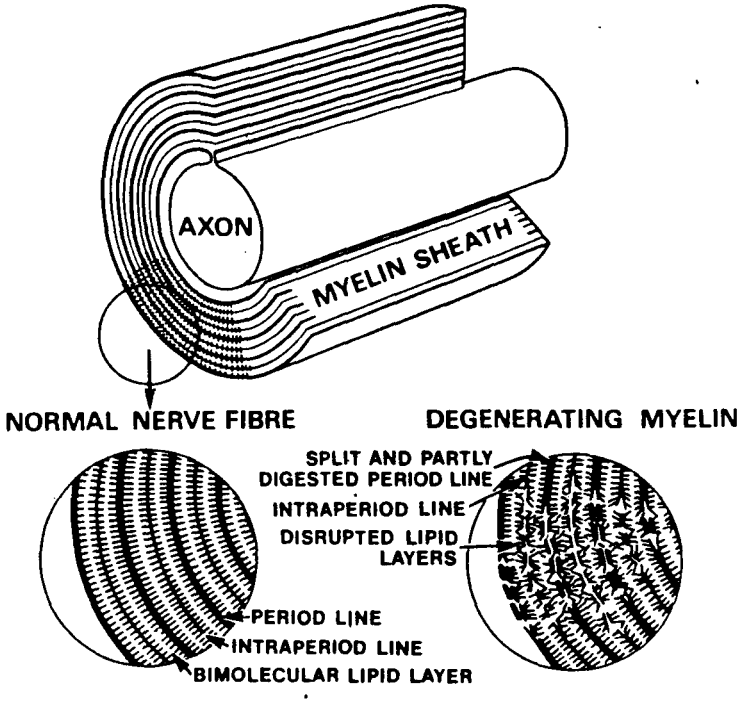

Fig. 8.20 Diagram of the effect of proteolytic enzymes on myelin structure, to show removal of protein lamellar components with resulting release of lipids. (Reproduced with permission from *Dev. Med. Child Neurol.*, 1962, 4, 393.)

basic protein leads to complete collapse of myelin, even though its function may be abnormal (Kirschner and Ganser, 1980).

It is of interest that the activity of α_1-trypsin inhibitor is moderately suppressed in the CSF in multiple sclerosis (Price and Cuzner, 1979), and that some success has been claimed for administration of a trypsin-inhibitor in experimental allergic encephalomyelitis in that it reduced clinical severity and the extent of demyelination (Boehme and Marks, 1978; Boehme *et al.*, 1978; Brosnan *et al.*, 1980). It is as though neutralization of a lysosomal enzyme protects myelin from its vulnerable position as an innocent bystander in a perivascular cell-mediated immune reaction (see Wisniewski and Bloom, 1975; Wisniewski, 1977).

It should not be forgotten that other lysosomal or plasma enzymes may also play a part in the demyelinating process. For example, lysolecithin solubilizes myelin (Webster, 1957) and induces demyelination (Gregson and Hall, 1973; Blakemore, 1978). Lysolecithin may be formed in the brain by the action of phospholipase A (Gallai-Hatchard *et al.*, 1972; Webster and Cooper, 1968) or in plasma by the enzyme LCAT (lecithin: cholesterol acyl transferase). Plasma lysolecithin is increased in multiple sclerosis and plasma LCAT is increased in acute episodes of the disease (Andreoli *et al.*, 1973).

Opinions differ as to the source of the increased lysosomal enzymes in and around the plaque of multiple sclerosis. It has been suggested that such enzymes are derived from proliferating neuroglia (see Adams, 1972), macrophages (Bower and Davison, 1974; Cammer *et al.*, 1978; Norton *et al.*, 1978) and from polymorphonuclear leucocytes (Cuzner, Davison and Rudge, 1978). However, as mentioned in Section 8.10, the mononuclear phagocytes in the plaque differ from monocytes and macrophages elsewhere in the body, while polymorphonuclear leucocytes are not commonly seen either in the lesion or in the CSF. Hence, work on non-neural phagocytic cells may not be relevant to the lesion. It could be argued that the lysosomal enzymes are derived from monocytes or macrophages in the perivenular inflammatory infiltrates but, against this, we have not so far obtained convincing histochemical evidence of active proteolysis in perivenular regions in the multiple sclerosis brain.

Arstila *et al.* (1973) considered that astrocytic lysosomes may be the source of these lysosomal enzymes, and this idea has been supported by Cuzner *et al.* (1976), McKeown and Allen (1978; 1980) and Allen *et al.* (1979). These last workers, using an azonaphthol histochemical method, identified marked acid phosphatase activity in grossly normal white matter in multiple sclerosis. Figure 8.21 illustrates acid phosphatase activity in the phagocytic cells of the plaque and weaker activity that is presumably in astrocytes in the surrounding white matter.

The gelatin film autogram method (Adams and Tuqan, 1961b; Adams, 1968) shows relatively little proteolytic activity in 'normal' white matter in multiple sclerosis and most is concentrated in the rim of proliferating

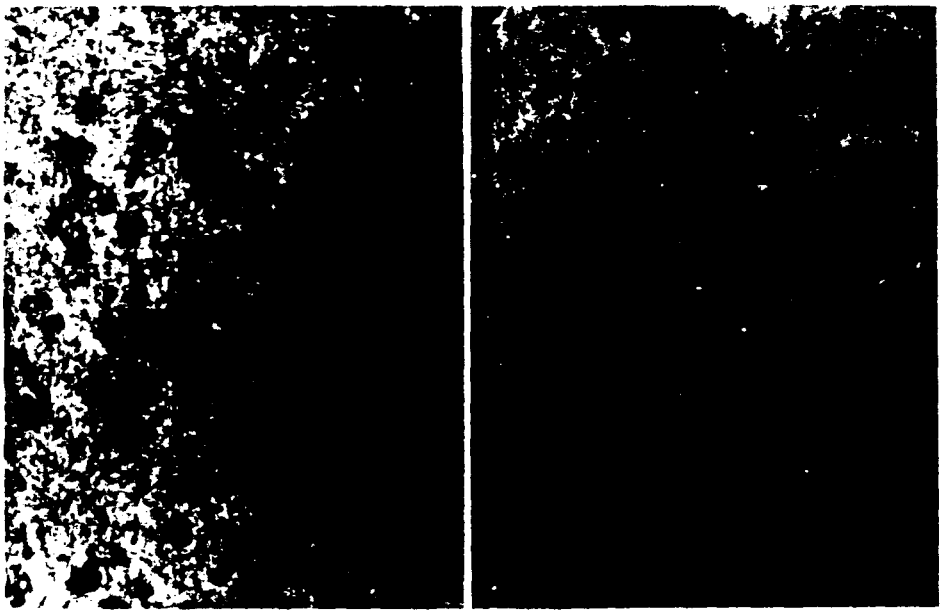

Fig. 8.21 Left: acid phosphatase activity mainly in lipid-laden mononuclear phagocytes at edge of a plaque of multiple sclerosis. Right: weaker reaction in presumed astrocytes in surrounding white matter. Hexazotized pararosaniline-naphthol-ASTR-phosphate. ×279.

neuroglia (Adams, 1968) observed at the edge of active plaques (see Friede, 1961, 1966; Friede and Knoller, 1964; Ibrahim and Adams, 1963, 1965). Thus, although some lysosomal enzyme activities can be attributed to astrocytes in surrounding white matter in multiple sclerosis, it is not clear whether this also applies to lysosomal proteases.

As discussed above, the lysosomal enzymes that increase in or around the multiple sclerosis plaque include acid proteinase, acid phosphatase (Hirsch *et al.*, 1976; Allen *et al.*, 1979) and various glycosidases (Cuzner *et al.*, 1976; McKeown and Allen, 1978). The increase in such enzymes is more likely to reflect the inflammatory nature of the lesion and, indeed, it would be most strange if lysosomal enzyme activity was not prominent in an inflammatory lesion (see Huszak, 1972).

In the broadest sense, all the cells discussed above could be considered to be reactants in inflammation. The identification of individual reactant lysosomal enzyme activities cannot help in resolving the nature of the aetiological agent in multiple sclerosis. At best this approach can explore the possibility of protecting myelin by stabilizing the lysosomal membranes of local inflammatory cells, either through suppression by steroids of an immune reaction or, as discussed above, by inhibition of the destructive effects of these lysosomal enzymes (e.g. with soybean trypsin inhibitor).

8.15 PATHOGENESIS OF MULTIPLE SCLEROSIS

In this chapter, it has been suggested that multiple sclerosis may result from the confluence of many minor demyelinating lesions initially caused, for example, by a viral perivenular encephalomyelitis (see Fig. 8.22). Because the inflammatory infiltrate in multiple sclerosis is often relative mild, this encephalomyelitis would have to be of a less acute or less severe grade than that encountered in a typical viral infection. The confluence of such small perivenular lesions might result from a local autoimmune reaction, much as is thought to occur in the localized lesion of adult-type post-primary pulmonary tuberculosis. However, it must be emphasized that the suggestions in this paragraph are absolutely speculative and there is (as yet) little concrete evidence for them.

Many of the pathological features of the lesion in multiple sclerosis indicate that the disease should be classified as a chronic progressive encephalomyelitis. The disease has in the past been enshrouded in pathological mystery by attaching to it names such as insular sclerosis, disseminated sclerosis and, more recently, multiple sclerosis. However, the only real pathological mystery—to leave aetiology aside—is the selective vulnerability of the myelin or, to turn the question on its head, the remarkable resistance of the axon.

A clinician might argue that the term progressive is inappropriate and that the word relapsing should be substituted. This would be justified by the natural history and clinical behaviour of the disease. However, it is not at all

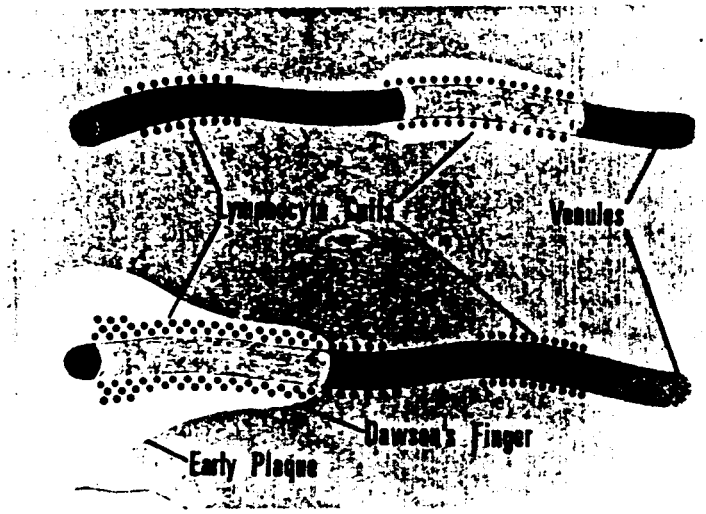

Fig. 8.22 Diagram to show perivenular lymphocytic infiltrates, and the formation of Dawson's fingers by extension around a venule. Coalescence of perivenular lesions may lead to the formation of a large lesion.

certain that an individual plaque undergoes cyclic exacerbation and relapse. If it has become inactive, microscopic appearances suggest that it probably remains so. Relapses are more likely to be due to progression of the disease near an existing plaque or elsewhere in the brain. Our observation is that many cases of multiple sclerosis show some areas of continuing active inflammation, even on occasions when the disease is considered to be clinically quiescent. If this observation is correct, then on pathological grounds the disease should be regarded as progressive and not relapsing.

As previously mentioned, the frequency of lesions near a ventricular surface and the continuity of lymphocytic and mononuclear infiltrates in the meningeal spaces with those in the perivenular Virchow-Robin space raises the possibility that something in the cerebrospinal fluid diffuses or spreads into the brain and triggers the lesion. It is quite unclear as to what this something might be. To suggest that meningeal lymphocytes start the process may be to put the hen before the egg. Nevertheless, the possibility should be born in mind that multiple sclerosis may start or progress as a lymphocytic meningitis, populated by what Lumsden (1970) termed immuno-agressive lymphoid cells. It could well be that an initial cell-mediated immune reaction, perhaps against a virus, occurs in the Virchow-Robin space or in the leptomeninges and that myelin is only involved as an incidental effect. Thus, the myelin sheath would only be damaged as a vulnerable innocent bystander (Wisniewski and Bloom, 1975; Wisniewski, 1977).

The role of linoleic acid in the pathogenesis of multiple sclerosis is quite unclear. It is possible, but unlikely, that a deficiency of this fatty acid causes an instability of the molecular structure of the myelin sheath and other membranes (see Caspary, Sewell and Field, 1967; Schauf, Frischer and Davis, 1980). It is perhaps more reasonable to emphasize the immunosuppressive action of this 18:2 fatty acid and to speculate on its role in prostaglandin production, which might also explain the increased platelet adhesiveness in patients with multiple sclerosis.

The histopathology of multiple sclerosis has produced a number of interesting avenues to explore but, nevertheless, it remains hazardous too strongly to identify any particular one as the likely cause. It is appropriate to end with Lumsden's (1970) warning that 'pathology is still the indispensable compass with which experimental and epidemiological research must be steered, and without which that research will be wasteful and meaningless.'

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CHAPTER NINE

The Possible Role of Viral Infections in Multiple Sclerosis and Other Related Demyelinating Diseases

Volker ter Meulen and John R. Stephenson

9.1 Introduction
9.2 Demyelinating diseases associated with a viral agent
9.3 Evidence for a possible viral aetiology in MS
9.4 Conclusions and interpretations

9.1 INTRODUCTION

An infectious agent as the cause of multiple sclerosis (MS) has always been an attractive hypothesis as documented by the medical history of this disease of the human central nervous system (CNS). The idea of an infectious CNS process was born 100 years ago (Marie, 1884) and had been recurrently considered as a plausible underlying pathogenetic mechanism for this disease. Almost every kind of microbe has been incriminated in the past (Johnson, 1975), but up to the present, no infectious agent has been isolated which could be aetiologically linked to MS. However, despite the failure to find a microbial agent, there is circumstantial evidence which still suggests that such an association could exist. Moreover, investigations of other human and animal CNS diseases have shown that viruses can induce demyelination, do persist over years in CNS tissue and may cause a chronic disease process different from the well-known viral induced acute infections (ter Meulen and Hall, 1978).

This chapter presents the evidence for and against the hypothesis of a viral aetiology in multiple sclerosis, as well as the virological aspects observed in other demyelinating diseases in animals and man. Since many reviews on this subject have been written in the past, only recent findings are considered and summarized in an attempt to discuss and interpret the mechanisms by which viruses could induce chronic CNS disorders associated with demyelination.

9.2 DEMYELINATING DISEASES ASSOCIATED WITH A VIRAL AGENT

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CHAPTER TEN

The Cerebrospinal Fluid in Multiple Sclerosis

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with the assistance of
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10.1 Introduction
10.2 Practical aspects of the cerebrospinal fluid (CSF) profile indicative of multiple sclerosis (MS)
10.3 Methodology
10.4 The central nervous system (CNS) as an immunological organ in MS
10.5 Conclusion

10.1 INTRODUCTION

The diagnosis of multiple sclerosis (MS) is clinical (Hallpike, Chapter 5). At the present time laboratory investigations cannot establish the diagnosis of MS, nor can they negate a diagnosis of clinically definite disease. Laboratory studies can add confidence to a diagnosis of clinically definite MS or contribute towards making the diagnosis of MS untenable.

The cardinal disturbance manifested by over 90% of patients with MS is IgG synthesis within the blood–brain barrier (BBB). The central nervous system (CNS) is not normally a repository for immunoglobulin (Ig) synthesizing cells but in MS patients there are both qualitative and quantitative abnormalities of CNS and cerebrospinal fluid (CSF) IgG. Once such IgG synthesis is detected in MS patients, it tends to persist for the duration of the patient's life and manifests great clonal restriction and stability of electrophoretic mobility over long periods of time. These Igs are heterogeneous in known specificities but the specificity of the largest portion of the Ig produced is unknown. The major portion of Ig synthesized intra-BBB displays considerable homogeneity as manifested by class, subclass, light chain, allotypic, and electrophoretic restriction. Such profiles are not common in other human diseases as far as is known. To some, this has suggested that IgG synthesis in MS is not due to a random activation of B cells but represents an immune response to a particular antigen or antigens in MS brain. Ig-producing cells have now been identified in MS brain and it is thought that the abnormalities of CSF IgG

(exacerbating, stationary, or remitting), except that production was reduced by administration of ACTH and/or steroid putative therapy (see Section 10.4.2 (f)).

Stendahl-Brodin and Link (1980) have attempted to relate the prognosis of MS with the presence of intra-BBB IgG synthesis as manifested by CSF oligoclonal IgG and elevated Link/Tibbling IgG index. Out of 17 MS patients without oligoclonal IgG banding 14 (82%) displayed slight or no disability after a mean duration of MS of 17 years while 53% of 88 patients with oligoclonal CSF IgG had a benign course after a mean duration of disease of 13 years. A benign course was also more often accompanied by a normal IgG index. Hershey and Trotter (1980) found that neither the patient's age, sex, duration of illness, activity of disease, nor longitudinal studies correlated with IgG/albumin, Link/Tibbling IgG index, the Tourtellotte empirical formula, agarose-gel electrophoresis and isoelectric focusing to detect oligoclonal bands. Similarly, no correlation between age, duration, clinical course and intra-BBB IgG synthesis (CSF oligoclonal IgG manifested by isoelectric focusing, Link/Tibbling IgG index and the Tourtellotte empirical formula) was found by Livrea *et al.* (1981).

In conclusion, the incidence of abnormal CSF findings increases with clinical confidence in diagnosis. About 90% of clinically definite MS patients have increased intra-BBB IgG synthesis manifested by a CSF IgG oligoclonal pattern, whereas only half of the patients classified as suspected MS (a single episode suggestive of the disease with or without evidence of a single lesion) have an oligoclonal pattern (Thompson, 1977). Furthermore, there is a tendency for a correlation between intra-BBB IgG synthesis and multiple attacks, multiple lesions and marked functional disability which accompanies confidence in the diagnosis. Accordingly, intra-BBB IgG synthesis in the MS CNS can be regarded as a barometer of ongoing destruction of the CNS myelin.

10.2.4 Differential diagnosis

Numerous reports (Harter, Yahr and Kabat, 1962; Bergmann and Gilland, 1968; Laterre *et al.*, 1970; Link and Muller, 1971; Thompson, 1977) have indicated that increased intra-BBB IgG synthesis can be caused by diseases other than MS. After excluding patients with marked pleocytosis (≥50 leukocytes), marked total protein elevation (≥100 mg dl^{-1}), positive syphilitic reaction, abnormal serum electrophoresis, and abnormal CSF dynamics, increase in intra-BBB IgG synthesis was found in the following conditions: MS, SSPE, chronic phases of CNS infection such as meningoencephalitis due to bacteria, parasites (toxoplasmosis, cysticercosis, trypanosomiasis), mycosis and viral infections, Guillain-Barré syndrome, cerebrovascular disease, brain tumours, and CNS vasculitides. A recent study by Ebers and Paty (1980) utilizing agarose and/or cellulose acetate for electrophoresis as a means to

detect oligoclonal IgG bands on 1000 patients found the following: in patients with clinically definite MS ($n = 267$) 92.8% had oligoclonal banding. In patients with possible MS ($n = 283$) oligoclonal banding was present in 31.1%. In patients with other neurological diseases ($n = 450$) oligoclonal banding was present in 8% ($n = 36$). Nineteen non-MS patients with positive oligoclonal banding had serum bands or disorders known to be associated with local CNS immune response. The remaining 17 patients had no explanation for the oligoclonal banding; in the majority of these patients, MS had not been a diagnostic consideration. They concluded that CSF electrophoresis to detect oligoclonal IgG bands is the single most reliable test in MS and deserves to be incorporated into the diagnostic criteria for this disease.

10.2.5 Conclusion

Clearly then, the MS CSF profile indicative of MS is not diagnostic. It is only useful to the clinician to support the diagnosis of MS. From an immunological as well as neuropathological standpoint, it is a response with features shared with other disease processes affecting the CNS. Accordingly, the physician must be alerted that evidence of the MS CNS profile indicative of MS is not unique to MS but is occasionally manifested in other CNS diseases.

10.3 METHODOLOGY

10.3.1 Introduction

As scientific reports were reviewed for this chapter, it became obvious that many conflicts in the literature were partly or principally due to differences in methodology by various investigators. For this reason, we felt it useful to state briefly some observations from our own experiences which might be helpful in the laboratory investigations of patients being evaluated for MS.

10.3.2 Quantitation of IgG and albumin

Serum is collected at the time of lumbar puncture. Several systemic protein abnormalities are reflected in CSF and if serum is not analysed simultaneously with CSF, errors may result. IgG and albumin can be measured concomitantly in CSF and serum by electroimmunodiffusion (EID) (Tourtellotte *et al.*, 1971b) using plates and antisera made in the laboratory (Fig. 10.1). Electroimmunodiffusion is a simple, rapid, and accurate method which requires only 5 μl of CSF. It is more sensitive than radial immunodiffusion (Schuller, Lefevre and Tompe, 1972) and has the additional advantage that albumin and IgG can be measured simultaneously on the same plate, thus

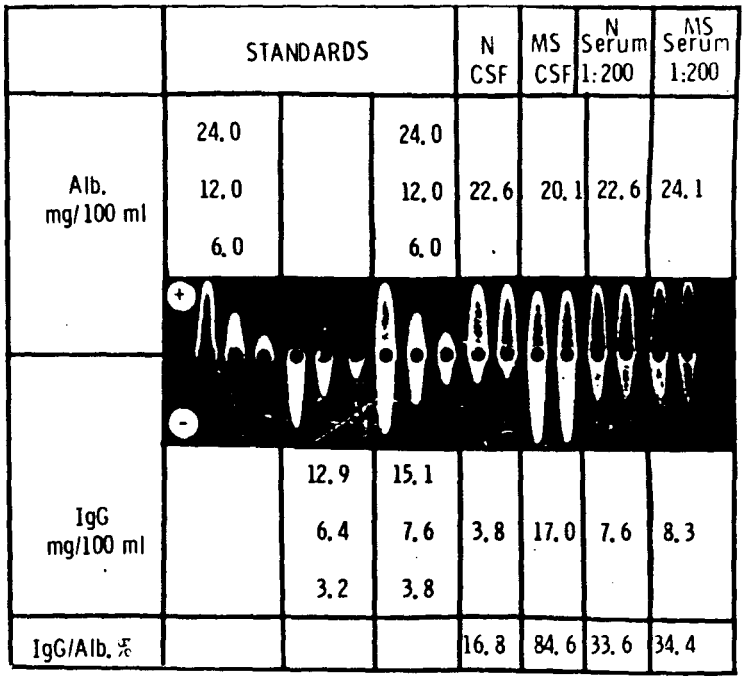

Fig. 10.1 Typical electroimmunodiffusion plate (Darkfield photograph). Application wells, middle of plate, contain 5 μl. Alb indicates albumin, MS, multiple sclerosis, N, normal. Reprinted with permission from Tourtellotte *et al.* (1971b).

permitting the quantitation of intra-BBB IgG synthesis in mg per day (see Section 10.4.2(e)). The laser nephelometer method is also reliable (Mandler, Goren and Valenzuala, 1981) and has the advantage of automation. However, if only a few specimens are to be measured each week the EID method which includes standards in each plate is more economical in cost and technician time.

10.3.3 Electrophoresis and isoelectric focusing

Electrophoresis of CSF has been carried out on a variety of media (cellulose acetate, agar, agarose, etc.). Cawley *et al.* (1976), Ma *et al.* (1977), Johnson *et al.* (1977), and Hershey and Trotter (1980) have used a commercially available method (Panagel, Worthington Laboratories, Freehold, NJ 07728, USA) for defining oligoclonal IgG bands which is suitable for the clinical laboratory (Fig. 10.2). It is simple to perform and sensitive. The disadvantage is that the sample must be concentrated to approximately 10–15 mg ml^{-1}. Ma *et al.* (1977) were able to avoid preliminary concentration in many CSF specimens by utilizing immunofixation. The technique entails application of 3 μl of CSF containing 1.0 μg of IgG, electrophoresis, and immunofixation by layering a

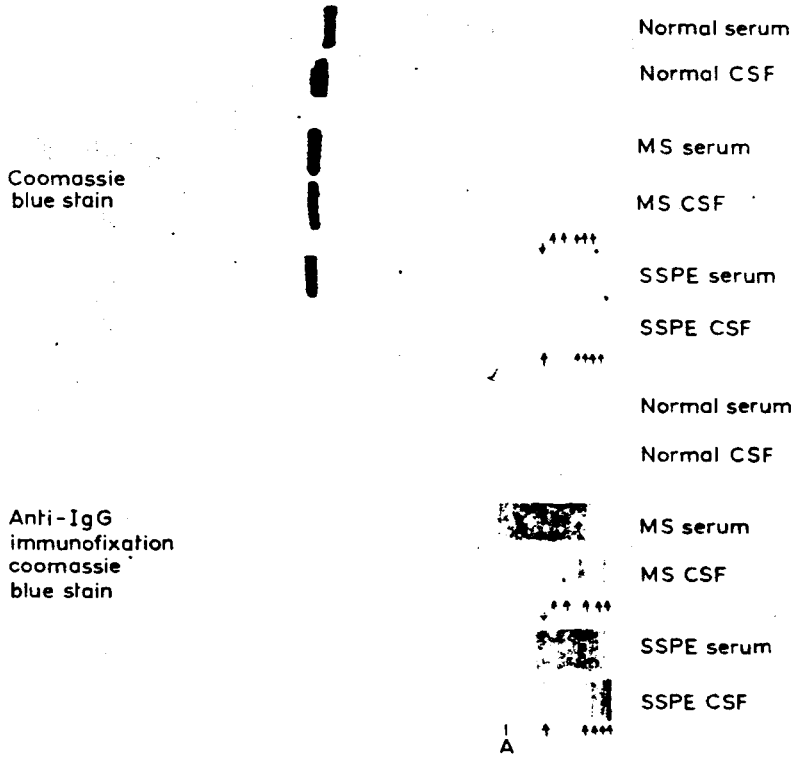

Fig. 10.2 Agarose electrophoresis (Panagel) of CSF and serum specimens. Five μg of IgG was applied to each lane followed by picric acid fixation and Coomassie blue stain and by IgG specific immunofixation and Coomassie blue stain (Cawley *et al.*, 1976). Arrows denote oligoclonal bands. 'A' marks the application site.

monospecific IgG antiserum over the separated proteins, followed by staining with Coomassie blue. Preliminary concentration of CSF is not required when the IgG concentration exceeds 10 mg dl^{-1}. In lieu of concentration of CSF, multiple applications of as many as five 3-μl samples of neat CSF, totalling 1 μg of IgG have been found to be equivalent to preliminary concentration. The addition of IgG immunofixation validates cathodic proteins as IgG and eliminates possible confusion with cathodic non-IgG proteins, such as δ and γ trace proteins. A standard amount of IgG should be added, for example 3 μl of fluid containing 1 μg of IgG, and immunofixation should always be used. Also a matched sample of serum should be diluted so that the same amount of IgG is added as that used for the CSF. If there is a corresponding IgG band in the serum to that in CSF, it is not possible to conclude that a CSF oligoclonal band is synthesized intra-BBB, i.e. the serum IgG could have transudated into the CSF and produced the band (Lumsden, 1972).

Several other methods have been described for identification of oligoclonal

bands as IgG by immunofixation (Ritchie and Smith, 1976; Arnaud *et al.*, 1977; Kahn and Thompson, 1978; Mattson, Roos and Arnason, 1980b) and the experimental details are provided in these reports.

Delmotte (1971) was the first to examine MS CSF by isoelectric focusing. Extensive experience has accumulated in the past few years. The method is exquisitely sensitive for the detection of oligoclonal IgG. The principal abnormalities of IgG are cathodic in location (Sidén, 1980). There are commercially available gels of high quality available complete with precise instructions. The method entails more expense than conventional electrophoresis and the abnormalities demonstrated are obviously more complex but sensitivity is somewhat better.

10.3.4 Concentration of CSF

It is convenient to use the Amicon CF15 concentrator (Amicon Corp., Lexington, Mass.) which is now very popular. However, there are drastic losses of protein including both IgG and albumin using this method. For more precise work, the use of the colloidion bag method of Kaplan and Johnstone (1966) gives approximately 90% recovery of IgG and albumin. No evidence has been provided that concentration *per se* alters the oligoclonal profile or leads to a preferential loss of some proteins.

10.3.5 Electrophoresis of unconcentrated CSF

Several methods are now available to demonstrate the presence of oligoclonal IgG in CSF without preliminary concentration. Some of these methods have been available for the past 10 years but their application to detection of CSF immunoglobulins in human neurological diseases has been delayed. In 1972, Kerenyi and Gallyas reported the use of silver staining to detect the fractionation of immunoglobulins in unconcentrated CSF from a patient with MS. Several silver stains have subsequently been popularized (Switzer, Merril and Shifrin, 1979; Merril, Dunau and Goldman, 1981a; Merril *et al.*, 1981b; Oakley, Kirsch and Morris, 1980) and several of these investigators have reported the use of silver stains to study CSF immunoglobulins. Allen (1980) has described a procedure for silver staining of isoelectric focusing polyacrylamide gels. Tourtellotte *et al.* have found that silver staining of MS CSF demonstrates excellent sensitivity for oligoclonal IgG when 1 μg of IgG is applied to the IEF gel lane. Immunofixation prior to silver staining increases the sensitivity of oligoclonal IgG detection (Fig. 10.3).

Cawley *et al.* (1976) and Ma *et al.* (1977) reported on several methods for the identification of oligoclonal IgG on unconcentrated CSF by the use of immunofixation methods with antiserum or antiserum conjugated to peroxidase. Oligoclones can only be detected by these methods if there is considerable elevation of CSF IgG to approximately 10–20 mg dl^{-1} which is

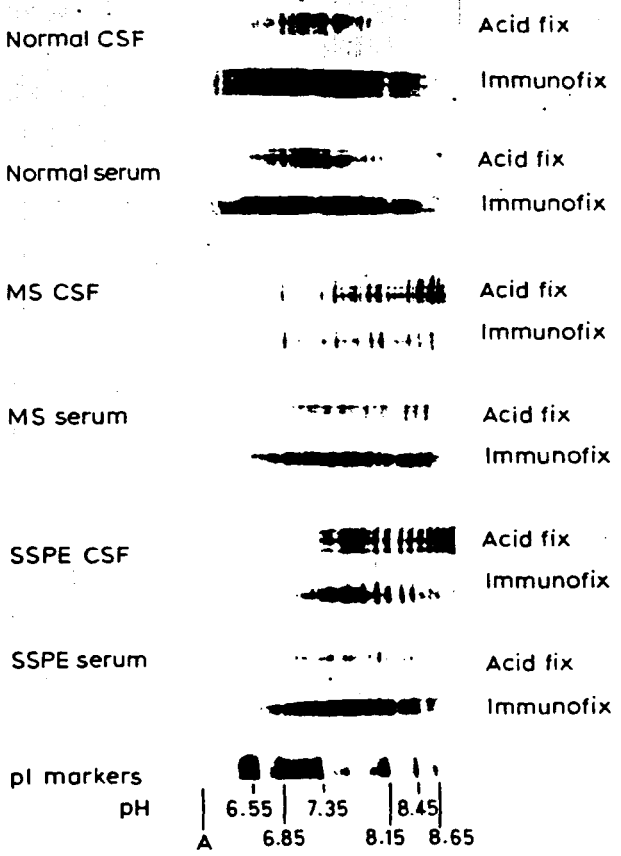

Fig. 10.3 Isoelectric focusing of CSF and serum specimens on LKB PAG-Plates (pH 3.5–9.5). In lanes marked acid fix two μg of IgG was applied, followed by fixation in trichloroacetic acid (11.5%) and sulphosalicylic acid (3.5%) solution and silver nitrate staining (Oakley *et al.*, 1980). Immunofix lanes demonstrate immunofixation with antisera to γ chains (Dako, diluted 1:10 of 1 μg IgG, followed by silver stain. 'A' denotes application site. Anodic half of gels are not shown.

substantially above the mean levels of IgG in MS patients. Mattson, Roos and Arnason (1980b) have described in detail a procedure for use of the peroxidase method but it suffers from the same disadvantage. Thompson *et al.* (1979) have obviated the need for CSF concentration by the use of acrylamide gel electrophoresis in vertical glass tubes which allows a large volume of sample (100–200 μl) to be applied to the gel prior to electrophoresis. Employing this method, excellent sensitivity in the detection of oligoclonal IgG in MS patients was reported. In 1978, Keck, Grossberg and Pressman reported the detection of Ig on polyacrylamide gels using radiolabelled antisera. Lasne *et al.* (1981) reported the use of radiodetection for CSF IgG with a sensitivity of 100 ng of IgG.

10.3.6 Polyacrylamide gel electrophoresis in sodium dodecyl sulphate (SDS-PAGE)

Carson *et al.* (1978) reported the use of SDS-PAGE to separate the components of CSF according to molecular weight. They then assayed gel slices for MBP and MBP-related activity by radioimmunoassay. Densitometric scans of gels from MS and non-MS patients were qualitatively similar although there were differences in some of the peaks between MS and non-MS patients. Hochberg and Wolfson (1979) also fractionated unconcentrated CSF in this way. They were able to demonstrate 'banding' in the gammaglobulin region in MS patients though no immunofixation data were reported. More recently, Iivanainen *et al.*(1981) studied 57 MS patients with SDS-PAGE using unconcentrated CSF and compared the results with agarose gel electrophoresis. A study was reported as positive if two or more bands were visible in the IgG region by SDS-PAGE using immunofixation. Of 57 patients 25 were positive using this criterion while the number of positive cases was only 18 of 57 MS patients using agarose gel electrophoresis, a lower figure than usually reported. SDS-PAGE also consistently revealed a larger number of bands in the IgG region than did agarose gel electrophoresis. SDS-PAGE separates molecules according to molecular weight. The heterogeneity demonstrated by this method then therefore presumably reflects molecular weight differences in the MS CSF IgGs. It is known that increasing pIs of IgG tend to be associated with increasing molecular weight. It is presumed, therefore, that the IgG fractionation demonstrated by SDS-PAGE is the molecular weight correlate of the cathodically migrating IgG seen using isoelectric focusing. The advantage of tube gel and vertical slab gel electrophoresis using either gradient electrophoresis or electrofocusing is that a large volume of sample such as 200 or 300 μl can be applied thus obviating the need for concentration. On the other hand, Iivanainen *et al.* (1981) have standardized a method requiring 50 μl of neat CSF. Silver staining of these gradient gels will even further enhance sensitivity.

10.4 THE CNS AS AN IMMUNOLOGICAL ORGAN IN MS

10.4.1 Introduction

The CSF often mirrors the extracellular environment of the CNS, therefore important aetiological, pathogenic, and therapeutic clues may be obtained from the study of CSF. The CSF not only fills the ventricles and surrounds the CNS but it also probably penetrates it by way of the Virchow-Robin spaces. Moreover, it is in close contact with but separated by a barrier from the blood

supply of the CNS as well as from the blood and lymphatic supply of its covering arachnoid membranes by cell to cell tight junctions. Tschirgi (1960) was one of the first investigators to present some evidence to support the hypothesis that the CSF is a pool of circulating fluid in communication with the ECF that surrounds the CNS cells; i.e. the CSF is an expansion of the extracellular space (ECS).

CSF is formed by the choroid plexus and circulates through the ventricles and in the subarachnoid space at the rate of 500 ml day^{-1} and non-specifically clears solutes from the ECS of the CNS; i.e. the CSF constitutes a low-concentration compartment (a 'sink') in widespread close contact with the CNS ECF, resulting in diffusion of solutes into the CSF (Rapaport, 1976). These solutes are then returned to the blood by bulk flow through the macropinocytosis system of the arachnoid villi. It might be said that the sink action of the CSF serves the function of a lymphatic drainage system adapted to the CNS (Oldendorf, 1967). The ependyma behaves like a loosely-knit membrane presumably with large gaps between its cells; moreover, according to Brightman (1965) it is permeable to large molecules, e.g. ferritin, 500 000 mol. wt. An easy exchange of solutes across the ependyma could be related to the composition of MS CSF, since Brownell and Hughes (1962) have shown that 40% of all the plaques of demyelination in chronic MS are located periventricularly. The extent to which Virchow-Robin spaces follow small vessels is not settled; if they penetrate to at least the arteriolar level, they could offer a potential communication between ECF and CSF in the subarachnoid space.

The size of the CNS ECS is about 20–30% of the volume of the brain and, thus, there is an adequate amount of ECS for the free diffusion of compounds in normal CNS. Electron micrographs of plaques of demyelinaton due to MS, obtained immediately after death, show an enlargement of the ECS when compared to the surrounding normal-appearing white matter (Prineas, 1975). Free movement of solutes the size of albumin and immunoglobulins between the CNS and the CSF, from the periventricular or surface plaques is therefore unlikely to be limited by the size of the ECS or by ependyma. Many MS plaques are deep in the white matter. Are these plaques accessible to the sink action of the CSF? Anatomical relationships of the ventricular system and subarachnoid space imply that no point of the CNS is more than 2 cm from the ependyma or pia (Oldendorf, 1967). Most regions are within 1 cm, and parts of the CNS most likely to have plaques (periventricular surface, surface of the brain stem and spinal cord) are within a few millimetres. Despite this proximity it is well recognized that lumbar CSF can be normal in patients who eventually turn out to have MS. A complete answer to this enigma cannot be given; it will have to wait until more is known about the disease and its relationship to the CSF. Perhaps the size of the lesion and/or the distance from the CSF space could still be a factor. Moreover, with 500 ml of CSF formed per day an abnormality could be diluted out.

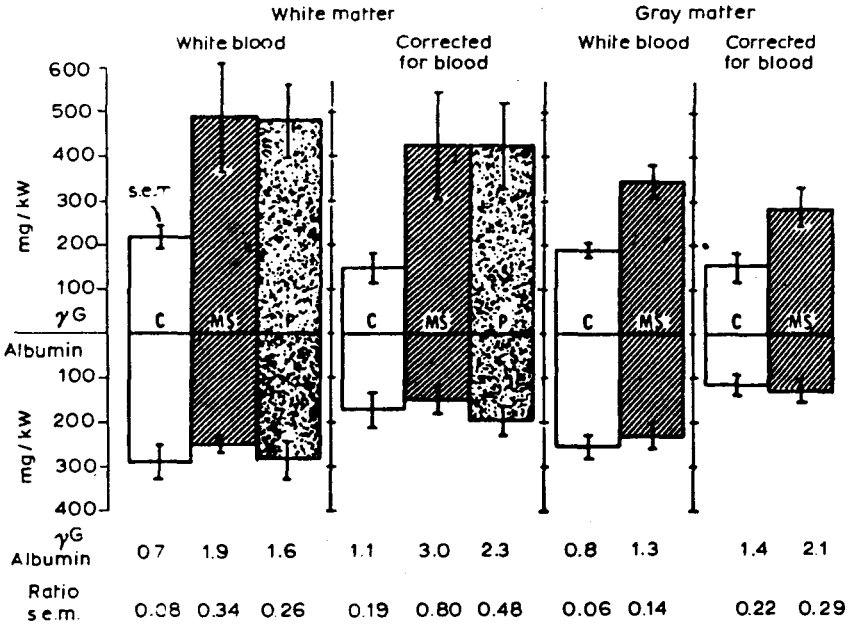

Fig. 10.4 Concentrations of IgG (γG) and albumin in post-mortem central nervous system (CNS) tissue from 10 control patients (C) and 11 multiple sclerosis (MS) patients. (P refers to plaque of demyelinated tissue.) Reprinted with permission from Tourtellotte and Parker (1967).

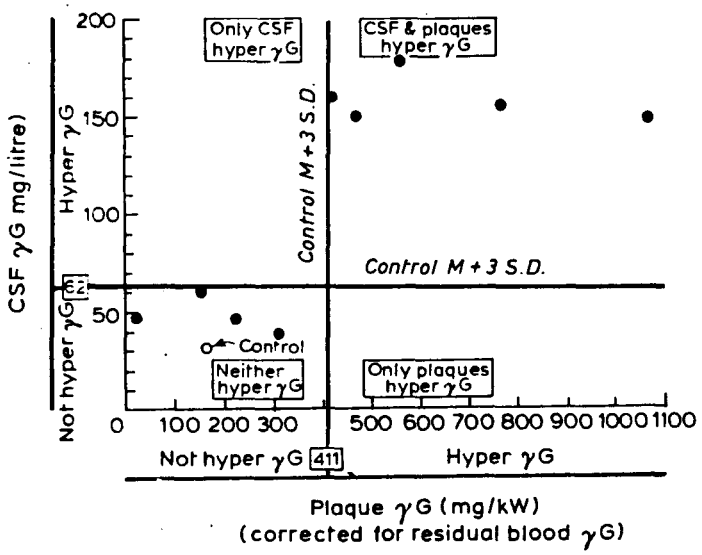

Fig. 10.5 Positive correlation between IgG (γG) in plaques of demyelination due to multiple sclerosis (MS) and that in the patient's cerebrospinal fluid (CSF) obtained before death. Reprinted with permission from Tourtellotte and Parker (1966).

Semiquantitative information on gamma globulin content of MS brain extracts provided by Link (1972b) resembled the earlier data of Tourtellotte and Parker (1966) and Tourtellotte, Itabashi and Parker (1967).

10.4.2(c) Electrophoresis of brain-extracted IgG

In 1972, Link (1972b) demonstrated two or three discrete oligoclonal bands in 6 of 10 hydrosoluble MS brain protein preparations. Significantly increased IgA was also found in these brain extracts. Mattson *et al.* (1980a) studied neutral and acid eluates from MS brain and control brain by isoelectric focusing and employed a direct peroxidase-conjugated antihuman IgG staining technique (Mattson *et al.*, 1980b). Normal saline and acid eluates from each of three plaques from a single brain had unique overall patterns of IgG bands despite many shared bands. The white matter pool acid eluate contained all the IgG bands found in the acid eluates from the individual plaques with additional bands. Saline eluates had more anodal and fewer cathodal bands than acid eluates. Comparable differences were seen in another MS brain studied. Saline eluates from three regions of an SSPE brain had identical patterns of oligoclonal IgG and similar findings in CSF. Acid eluates of three SSPE brain regions had also virtually identical oligoclonal IgG patterns. Mehta *et al.* (1981) have reported similar results.

The principle feature of these electrophoretic studies (Mattson *et al.*, 1980a) is the marked similarity of the banding pattern found in the MS brains studied. Repeated homogenization and acid elution may be expected to release intracellular IgG with charge differences reflecting post-translational alterations rather than primary sequence or variable region differences. Mattson *et al.* (1980b) also carried out preliminary fixation of the focused proteins prior to immunofixation which might result in altered reactivity between the focused IgG and antisera. Hence, additional bands in acid eluates of MS white matter need not necessarily be taken as evidence for the existence of separate antigens.

10.4.2(d) Radioactive IgG and albumin blood and CSF exchange studies in MS

The studies by Frick and Scheid-Seydel in 1958 provided new insight into the source of the elevated CSF IgG in MS. In their initial study (1958a), radioiodinated albumin was injected intravenously into 13 patients with neurological disorders including six patients with MS. Specific radioactivity was determined in CSF and serum samples were collected simultaneously at intervals for up to about 2 weeks after injection. The albumin passed quickly into the CSF compartment and equilibrium between CSF and serum was established at 60 h. A similar study (Frick and Scheid-Seydel, 1958b), but this time using radiolabelled IgG, was carried out in 28 patients with various neurological disorders, five of whom had MS. As with albumin, a steady-state equilibrium was established but required 100, rather than 60 h as for

quantity of IgG known to be produced each day within the CNS of affected patients. This was based on the average daily synthesis of myeloma cells *in vitro* and is only an approximation because of the large number of variables relating to IgG synthesis in any regional compartment of the immune system (Tourtellotte, 1970a). A special study of the leptomeninges was not performed in the study by Prineas and Wright (1978) although a prominent leptomeningitis may be seen in MS and is a potential source for some of the IgG produced *de novo* in MS (Adams, 1977).

10.4.8(a) The macrophage and the immune response in MS

Monocytes and macrophages compose a family of phagocytic cells. These cells are widely scattered throughout the body and include the alveolar macrophage, the Kupffer cell of the liver, the lining macrophages of the splenic and bone marrow sinusoids, and the freely migrating macrophages of the pleural and peritoneal cavity (reviewed by Cline *et al.*, 1978). The brain also possesses a cell population with similar functions. These cells have been known as microglia since the beginning of the century; their cellular origin is still in dispute but functionally they resemble systemic mononuclear phagocytes and have some morphological, biochemical and cell surface immune marker similarities (reviewed by Oehmichen, 1978).

There are a number of reasons for investigating the monocyte-macrophage cell series in human demyelinating diseases, specifically MS.

(1) In experimental demyelinating encephalopathies, the macrophage or cells with macrophage-like characteristics are seen actively stripping the myelin sheath (Wisniewski, 1977). In several experimental demyelinating encephalopathies, such as corona virus induced subacute demyelinating encephalomyelitis in rats (Nagashima *et al.*, 1978), Theiler's virus infection in mice (Penny and Wolinsky, 1979), and in canine distemper demyelinating encephalomyelitis (Summers, Greison and Appel, 1979), the macrophage is a prominent cellular component of the acute lesion and is believed to play an important part in the digestion and disposal of the degenerated myelin membrane and the other cellular components destroyed in the pathologic process.

(2) Enzyme inhibitors, especially inhibitors of mononuclear phagocyte proteolytic enzymes are modestly effective in the modulation or suppression of EAE (Cammer *et al.*, 1978; Sibley, Kiernat and Laguna, 1978; Bloom *et al.*, 1978).

(3) Enzyme assays have demonstrated alterations in levels of mononuclear phagocytic enzymes in plaques, periplaques and grossly unaffected white matter in MS. In addition, alterations in CSF levels of these enzymes and enzyme levels in circulating leukocytes have been documented to occur in MS (Cuzner, Davison and Rudge, 1978; Hallpike, Chapter 11; Adams, Chapter 8).

(4) Histological studies especially electron microscopy have identified the

macrophage as the most prominent cellular representative of the immune system in MS brain (Prineas and Wright, 1978). Other authors have commented on the significance of macrophages in MS lesions (Lumsden, 1972; Raine, 1978) and in MS CSF sediment studied by electron microscopy (Herndon and Kasckow, 1978).

15 Supernatants of activated mouse macrophages can digest MBP *in vitro*. This was demonstrated using polyacrylamide-gel electrophoresis prior and subsequent to incubation of the MBP with macrophage supernatants (Cammer *et al.*, 1978). Studies using enzyme inhibitors have implicated a plasminogen activator which acts on plasminogen, a protease precursor found in blood and body fluids. The enzyme has trypsin-like specificity and is a key enzyme in the clotting system, fibrinolysis and the complement cascade (Bloom *et al.*, 1978).

Prineas (1975) and Prineas and Wright (1978) studied the cellular pathology of MS brain. In one study (Prineas, 1975), perivascular macrophages with prominent cytoplasmic granules were one of the most common and easily recognizable cells seen within the perivascular compartment. Occasionally, the cells were filled with lipid vacuoles. On electron microscopy, the spherical cytoplasmic granules, which were often 2 μm or more in diameter, appeared to be unusually large but otherwise typical, membrane-bound primary lysosomes. Their structure generally was identical with free macrophages elsewhere in the body. Prineas has identified another 'microglial' cell type with phagocytic capacity in MS lesions (Prineas, 1975). This cell differed from the perivascular macrophage in a number of respects. First, large primary lysosomes were never observed in cells outside the perivascular compartment; second, characteristic inclusions consisting of membrane-bound stacks of curved, linear profiles were observed in almost virtually every microglial cell seen in demyelinated tissue but were rarely observed in perivascular macrophages, even when microglia filled with this material were seen clustered immediately outside the perivascular space; and third, the nucleus of a typical microglial cell was smaller, more irregular in shape and had more peripheral heterochromatin than the nucleus of a perivascular macrophage. Prineas felt that his observations supported previous impressions that microglia in MS plaques seemed unable to completely digest myelin, thus leading to the accumulation of distinctive cytoplasmic inclusions. In contrast, this material appears to be rapidly cleared from the perivascular compartment by perivascular macrophages which in contrast to microglia, contain very large primary lysosomes. Prineas felt that the microglial cells were derived entirely from locally proliferating cells which are not, or only slowly, exchanged with haematogenous cells. Prineas (1975) had earlier suggested that these cells might in fact be oligodendrocytes and that demyelination in MS may involve intracellular absorption by oligodendrocytes of their own myelin to form fat-laden cells. Alternatively, they may represent the classic microglial cell.

Prineas and Graham (1981) also examined macrophages for IgG and

albumin in actively demyelinating lesions in two patients with MS using the peroxidase–antiperoxidase immunocytochemical technique. In both cases, macrophages were present that stained for cytoplasmic or surface IgG or both. In one case, in which the tissue was rapidly fixed in chilled fixative, macrophages located among myelinated nerve fibres at plaque margins, but not elsewhere in the plaque, revealed surface IgG in the form of caps restricted to one or both poles of the cell. These caps were absent in sections stained for albumin. Because capping implies the presence of a multivalent ligand close to the cell surface and because cap formation was observed only in macrophages contacting myelin sheaths, they suggested that antimyelin antibody cytophilic for macrophages may be present in the CNS in MS, and that immune ligand-mediated phagocytosis may play a role in myelin breakdown in the disease. This study provides the first direct evidence that IgG participates locally in myelin breakdown in MS. They concluded that the IgG caps may represent antibody cytophilic for macrophages (defining cytophilic antibodies as opsonins for which specific receptors exist on macrophage surface membrane) directed against an antigen expressed in plaque margins and associated with myelin sheaths. As only the IgG1 and IgG3 subclasses of IgG are cytophilic for human macrophages and monocytes (Lawrence, Weigle and Spiegelberg, 1975), it should be mentioned that the oligoclonal IgG of the CSF and brain in MS belongs mainly to the IgG1 subclass (see Section 10.4.4(b)).

10.4.8(b) Lymphatic channels in the CNS

Although a formal lymphatic drainage apparatus does not exist in the CNS, there does exist a system of non-endothelialized spaces, and potential spaces, lying adjacent to the basement membrane of capillaries and in the adventitia of the larger vessels. They pass from the depths of the cerebral cortex to discharge their contents into the cervical lymphatics of the neck. Casley-Smith (1976) has used the term 'prelymphatic' for this system. Prineas (1979) has studied this system in patients with neurological diseases including three patients with MS, using electron microscopy. Histology in the cases studied revealed the presence of lymphocytes and macrophages in the perivascular spaces of histologically normal white matter. Plasma cells were also seen in the MS patients. These three cell types were not randomly distributed within the perivascular spaces; the lymphocytes and macrophages tended to be confined within thin-walled channels, whereas any plasma cells present were distributed outside these channels. As compared with the channels in normal white matter, the channels associated with plaques were more numerous and more irregular in shape, and they were separated by collagenous trabeculae which contained isolated plasma cells or groups of plasma cells clustered around cells of the same type as those that formed the walls of the channels. Intimate contact was also observed between lymphocytes and macrophages inside the thin-walled channels observed in the perivascular spaces in each of

contained 10 or more leukocytes per mm³ had an average IgG value of 0.22. Furthermore, the MS patients treated by Tourtellotte in the cooperative study (Rose *et al.*, 1970) also showed a direct relationship between the IgG value and the total leukocyte count. Six patients at their initial CSF examination, all in relapse, had an IgG value of $\leqslant 0.15$; they had an average leukocyte count of 5 per mm³ (range 1–14). The 19 patients who had an IgG value >0.15 had a leukocyte count of 8 (range 1–30). At the time of a second CSF examination, approximately 1-year interval, seven patients declared a relapse and 15 a remission. Two patients who had a relapse and who had a CSF IgG value $\leqslant 0.15$ had four leukocytes (1 and 6) and the other five patients who declared a relapse and who had a CSF IgG value >0.15 had 9 cells (range 7–21). The three patients who had a remission and who had a CSF IgG value $\leqslant 0.15$ had four leukocytes (range 1–6) and the other 15 patients in remission and who had a CSF IgG value >0.15 had 9 (range 2–26).

Thompson (1977) pointed out that there has been no systematic study of the relation between electrophoretic patterns and the differential cytology of CSF in patients with MS. Since MS CSF lymphocytes have been shown to synthesize oligoclonal IgG it was of interest that all 22 patients reported with atypical lymphocytes or plasma cells showed oligoclonal bands whether pleocytosis was present or not.

10.4.9(c) CSF and CNS leukocytes and intra-BBB IgG synthesis

Regardless of the precise correlation between pleocytosis and elevation of gamma globulin in the CSF, the question arises as to the capability of CSF leukocytes to synthesize gamma globulin. Hirschhorn *et al.* (1963) calculated that 1 million lymphocytes stimulated by phytohaemagglutinin produced 10 μg of IgG in 24 h. This result is comparable with the findings of Salmon (1973) who has quantitated *in vitro* synthesis of IgG by myeloma cells and found average values of 10–20 pg of IgG per cell synthesized per 24 h. The average MS patient has 10 mg of IgG per 100 ml of CSF. A fraction of this IgG originates from the blood. If no damage to the BBB is assumed, it is equal to that amount found in normal CSF which is 3.6 mg dl^{-1}. Hence, 6 mg of IgG is accounted for by intra-BBB synthesis. If 500 ml of CSF is formed per day (Cutler *et al.*, 1968) then 30 mg of IgG is synthesized per day (Tourtellotte, 1970a; Tourtellotte and Ma, 1978). The average MS patient has six leukocytes per mm³ of which 90% are lymphocytes; hence, it turns out that there are on the average 5.4 lymphocytes per mm³ or 5400 lymphocytes per ml or $2.7 \cdot 10^6$ lymphocytes in the circulating CSF per day (500 ml of CSF turned over per day). A million lymphocytes maximally stimulated can produce 10 μg of IgG per day according to Forbes and Henderson (1966). Hence 27 μg could be synthesized by the lymphocytes which appeared in the circulating CSF in a day or <0.1% of the 32 mg to be accounted for. On the other hand, if lymphocytes in the CNS are responsible for the remainder of the CSF IgG in MS, 3.2 billion lymphocytes would be needed. This estimate was commented on by

manifestations since damage to the patient's CNS by demyelination might occur in the absence of clinical symptoms. Second. it is unknown whether the alteration of this cell subpopulaton represents a preferential migration of lymphocytes to the CNS in relation to disease relapse or peripheral destruction by one of several mechanisms. Third, is this alteration beneficial for or destructive to the patient's CNS? Fourth, is the phenomenon primary or a secondary pathogenically insignificant reaction to some other general disease-inducing process? MS patients in the absence of neurogenic involvement of lungs and bladder are generally in excellent health systemically. Unlike other disorders with a presumed autoimmune pathogenesis there is no obvious tendency so far reported for an increased incidence of systemic or extra-neural diseases. This suggests that the primary disease-inducing process is in the CNS and not in the bone marrow or elsewhere in the body. There is substantial evidence, however, that genetic factors are also operative in the establishment of disease and ongoing cellular studies should help identify these factors. The extent of variability in the reports relating to subpopulations of immune cells in CSF and blood as well as the lack of unanimity among investigators on the functional aspects of cell populations should be adequate to indicate the pressing need for standardized methods for the investigation of CSF cells and their functional classification.

10.5 CONCLUSION

We have selected from the very large literature to highlight the principal and most characteristic findings seen in MS CSF. The principal abnormalities on which there is a concensus reflect what appears to be persuasive evidence for an active immune process intra-BBB in MS patients which results in destruction of the myelin sheath with variable clinical consequences. The rate of synthesis of IgG intra-BBB and the CSF oligoclonal IgG response as a manifestation of intra-BBB IgG synthesis stand out as the most consistent and characteristic aspect of the CSF profile of MS. Many other laboratory abnormalities occur but few occur so regularly. It is now possible to modulate IgG synthesis. It might also be possible to eradicate IgG synthesis and the oligoclonal IgG response by some forms of immunosuppression. *Unfortunately, we do not yet know if IgG synthesis intra-BBB is epiphenomenal or an important cause of the pathology seen in MS.* The best indices now available correlate only poorly with disease activity. If the findings at autopsy or the results of neurophysiological studies such as evoked potentials, with or without provocative manoeuvres, and radiological studies utilizing CT brain scans (Sears, Hayman and Bigelow, 1981) are to be believed, much of the demyelination of the CNS in MS is entirely subclinical and it is perhaps therefore surprising that any laboratory index should correlate with disease activity as assessed clinically. We feel that what is needed is not so much laboratory indices of clinical activity but indices of pathological demyelination in MS CNS which are

subclinical. This information in turn may be of use as a scientific barometer for the effectiveness of putative therapies. Viral and autoimmune models of human demyelination should continue to be investigated with this goal in mind.

It is certain that a fully integrated view of the myriad changes in immune function in MS CNS and CSF is not possible at this time. Intra-BBB IgG synthesis is almost a *sine qua non* of MS. Cytology demonstrates the presence of large numbers of lymphocytes and macrophages colonizing the CNS in MS. Their considerable functional capabilities have been demonstrated in many ways *in vitro*. MS lesions are accompanied by BBB disturbance. Thus, the CNS, an immunologically privileged site begins to look like a lymphatic tissue. As a consequence of this and focal alterations in the BBB, the systemic immune system can impact and interact with the CNS. The humoral immune response is very diverse as indicated by the multitude of known specificities for MS CSF IgG. Quantitatively, however, the known specificities constitute apparently only a very small portion of total MS CSF IgG. By definition the range of antibodies demonstrated justify the characterization of the intra-BBB barrier humoral response as polyclonal B cell activation. Side by side with polyclonally synthesizing B cells having diverse antigenic specificities is the presence of IgG which demonstrates remarkable restrictions with considerable subclass, light chain, allotypic, and electrophoretic homogeneity. This response demonstrates great clonal stability and persists, once identified, for the duration of MS. These restricted Igs may be synthesized against MS antigen(s) and thus may represent powerful probes to identify the cause of MS. Faith, not objective evidence, however, is the main support for this position. It may be that humoral immune response in MS does indeed represent 'nonsense antibodies', secondary to random activation of B cells, unrelated to the cause of MS, and carrying no clues to its pathogenesis. However, if Prineas and Graham (1981) are correct about the capping of surface IgG, the predominant class for MS intra-BBB Ig synthesis, on macrophages engaged in myelin breakdown, there may be a pointer to the location of both antibody and antigen in MS lesions. Application of the increasingly powerful methods of molecular biology may resolve these questions in this decade.

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CHAPTER ELEVEN

Chemical Pathology of Multiple Sclerosis

J. F. Hallpike

11.1 Introduction
11.2 Protein changes
11.3 Lipid changes
11.4 Fatty acids
11.5 Platelet stickiness
11.6 Enzyme changes
11.7 Myelin basic protein in CSF
11.8 Immune complexes and T cell ratios
11.9 Amino acids

11.1 INTRODUCTION

Biochemical studies in patients with multiple sclerosis (MS) have largely been restricted to blood and cerebrospinal fluid (CSF), and brain obtained at autopsy. Their purpose has been to attempt to elucidate the disease process, to denote indices of disease activity and to aid diagnosis. Progress has been made in identifying certain consistent immunological responses in MS, such as the IgG changes and oligoclonal pattern in CSF described in detail in the previous chapter. Other data, which are negative or of no direct relevance, nevertheless consistute a perspective of the endeavours to resolve the enigma of this disease. Facets of information, for instance with respect to fatty acid metabolism, are intriguing and may yet have a place in the final scheme of pathogenesis. Tissue damage in MS is confined to the central nervous system (CNS) and the scope for direct investigation of the pathological process is correspondingly limited. In addition, the clinical expression of MS is characteristically variable in the early stages and systemic complications of more advanced disease serve to cloud interpretation of later biochemical findings. Immunological and cellular profiles in the CSF, histocompatibility (HLA) antigens and virological evidence in MS are considered in detail elsewhere in the book. The aim of this chapter is to review predominantly biochemical data in MS which continue to hold currency and are of potential significance for a better understanding of the condition.

11.2 PROTEIN CHANGES

11.2.1 Myelin

The essential pathological feature of the demyelinating process in MS is loss of myelin with relative preservation of axons. Whether such demyelination reflects intrinsic disease of the myelin sheath or arises through primary oligodendroglial injury is still unresolved. Breakdown of myelin may follow damage to its protein framework resulting in loss of lipid and of the radial stability of the sheath. Because of its encephalitogenic properties, the myelin basic protein (MBP) is probably the most fully investigated of all brain proteins (Shooter and Einstein, 1971; Carnegie and Moore, 1980). Loss or reductions of MBP in and around MS plaques (Hallpike and Adams, 1969; Hallpike *et al.*, 1970; Riekkinen *et al.*, 1971; Einstein, Dalal and Csejtey, 1970; Einstein *et al.*, 1972) are consistent with selective vulnerability of this myelin component to proteolytic attack (Adams and Bayliss, 1968; Csejtey *et al.*, 1972; Wood, Dawson and Hauser, 1974). Evidence of increased proteinase activity in cellular or 'active' MS lesions was reviewed by Hallpike (1972). These enzymes largely derive from or reflect the presence of macrophages (Norton *et al.*, 1978) although myelin itself has been shown to possess limited protease activity (Riekkinen and Clausen, 1969). While it is attractive to cast MBP as the target of an immune response in MS, with tissue damage from inflammatory mediators, such mechansisms have not been established as being of primary importance in the human disease.

The predominant protein in CNS myelin is proteolipid (Folch and Lees, 1951) which is resistant to proteolysis *in vitro* and which appears to retain its normal identity in early myelin breakdown. This myelin protein is, however, found to be degraded in established plaques (Csejtey *et al.*, 1975). Other minor protein components of myelin: Wolfgram protein, glycoproteins and DM-20 protein (see Norton, 1977a and Gregson, Chapter 1) have not been specifically studied in plaques or MS myelin. Glial fibrillary acidic (GFA) protein, a component of glial filaments, is present in MS plaques and appears to be a useful index of astrocyte reactions and gliosis in pathological tissue (Eng *et al.*, 1971; Eng, 1980).

11.2.2 Cerebrospinal fluid

Examination of the cerebrospinal fluid (CSF) by lumbar puncture is a valuable means of obtaining information of diagnostic significance in MS. As brain tissue can only very rarely be obtained in the patient's life time, study of the CSF is a requirement in seeking clues to the aetiopathogenesis of the disease. Added importance to spinal fluid analysis is provided by the proximity to the CSF pathway of sites of predilection for plaque formation, such as the periventricular region and surface white matter of the spinal cord. Information gathered from the study of cerebrospinal fluid proteins in MS

closely reflects the methodology employed. The importance of techniques was emphasized by Lumsden (1972) who vividly described the evolution in the last 40 years from measurements of total protein, to chemical determination of gammaglobulin, the introduction of electrophoretic methods and of immunochemical analysis. Total protein values in the CSF are within the normal range in at least 80% of cases of MS (Lowenthal, 1977), with elevations, when present, being usually slight and with values in excess of 1.0 g litre^{-1} occurring in approximately 1% of cases. Findings with CSF electrophoresis have varied in minor respects due to methodology and different ways of expressing results. In general, albumin, α- and β-globulin fractions are normal in MS, irrespective of duration of illness. Pre-albumin may be reduced (Clausen and Fog, 1969). Increases in α_2-macroglobulin and haptoglobulin occurring late in the disease are likely to reflect serum elevations of these proteins due to systemic complications. Haptoglobulin and glycoprotein increased in CSF in acute phases are more likely to be due to transient alterations of blood–CSF permeability for these proteins (Bauer and Gottesleben, 1969). Transferrins (Fe-binding proteins) show reductions in chronic cases, consistent with neuraminidase or proteolytic effects (Felgenhauer, 1971; Verheecke, 1975). A number of brain-specific proteins have now been assayed in CSF in demyelinating and other neurological diseases, in the hope of obtaining diagnostic information and also of identifying putative antigens. The glial S-100 protein is found in the CSF of patients with MS but is likely to be a non-specific accompaniment of parenchymatous disease (Michetti, Massaro and Murazio, 1979). The GFA protein can be measured in CSF (Eng, Lee and Miles, 1975) but data from longitudinal studies in MS are not yet available. Beta-trace protein, also thought to be of glial cell origin, is increased in CSF from severely disabled MS patients and probably reflects CNS tissue damage (Olsson, Link and Müller, 1976).

Present interest in proteins in the CSF centres on the increase in gammaglobulins. The original quantitative studies of gammaglobulin by Kabat and others, reviewed by Lumsden (1972), showed the CSF gammaglobulin to be increased above 13% of total protein in at least 80% of patients with MS, these values corresponding closely with the IgG 'index' (see Chapter 10) used currently. Electrophoretic studies reveal qualitative as well as quantitative changes in the distribution of CSF gammaglobulins. Extra electrophoretic bands visualized in the gamma region represent an oligoclonal pattern or reaction and are reported in over 90% of patients with clinically definite or probable MS (Thompson *et al.*, 1979). Using high resolution isoelectric focusing, oligoclonal bands were detected in 95% of patients with clinically verified MS and 30% of subjects with optic neuritis (Sidén and Kjellin, 1978). Immunoglobulins, i.e. proteins of plasma cell or B-lymphocyte origin which react directly with antigens, run mainly as gammaglobulins on electrophoresis but are identified immunochemically.

The raised CSF gammaglobulin in MS is almost entirely due to an increase in IgG, some of which is oligoclonal or of 'restricted heterogeneity' while the rest is diffuse or polyclonal. IgG subgroups have been demonstrated by radioimmunoassay and there appears to be a selective increase in IgG_1 in MS which correlates most closely with the total IgG concentration (Eickhoff *et al.*, 1979). Raised levels of IgM have also been found in MS (Williams *et al.*, 1978) but, in general, studies of IgA, IgM and IgE have not shown any specific or disease-related changes. Unlike subacute sclerosing panencephalitis (SSPE) in which condition high levels of measles-specific homogeneous IgG antibody occur in serum and CSF, the IgG in MS shows no consistent reactivity with measles (Haire, 1977; see also Chapter 9). Antbodies of the IgG class have been shown to react with a number of brain antigens associated with myelin, including cerebroside, sulphatide and non-lipid haptens (Ryberg, 1978). Other studies in serum failed to reveal glycolipid antibodies in MS (Leibowitz, 1980).

Other properties of CSF antibodies in MS have been studied. It was shown that IgG is produced in culture by lymphocytes obtained from CSF (Cohen and Bannister, 1967). Such IgG from CSF cell cultures is oligoclonal and synthesis of antibody is increased in exacerbations of the disease (Sandberg-Wollheim, 1974). Increased kappa:lambda ratio for light chains of IgG from CSF have been found in some 50% of MS patients (Link and Zettervall, 1970) with additional evidence from radial immunodiffusion of free kappa chains (Bauer, 1975). These subtle IgG changes are restricted to the CNS/CSF compartment. Immunoglobulin increases in serum in MS are homogeneously distributed and kappa:lambda ratios of serum IgG are normal. All these features constitute the 'CSF protein profile' in MS (Tourtellotte, 1975, Chapter 10) and are also indicative of synthesis of IgG in this disease within the CNS/CSF compartment (Ewan and Lachman, 1979). Although characteristic, increased CSF IgG and oligoclonal banding are not pathognomonic of MS, such changes being found in other neurological conditions including neurosyphilis, Guillain-Barré syndrome and some types of panencephalitis (Thompson, 1977) and acute cerebrovascular disease (Roström and Link, 1981).

11.2.3 Blood

Serum protein studies in MS, reviewed by Zilkha (1970), have shown reductions in albumin and elevations of globulins in the more severely affected and chronically ill. Such changes are interpreted as being non-specific and secondary to intercurrent infection or inanition. Quantitative immuno-electrophoretic studies (Clarke, Freeman and Pryse-Phillips, 1970) provided no evidence of MS-specific serum protein changes or of alterations related to duration of the disease. No differences in serum immunoglobulins (IgA, IgM, IgG) were found in MS versus non-neurological controls matched for age and sex (McMichael, Ross and Lenman, 1972). A further report (Delmotte and

Demonty, 1976) comparing 772 MS patients with 226 neurological controls, grouped according to age and sex, revealed only a slight increase in IgM in the MS group. Minor degrees of hypocomplementaemia have been found in MS correlated with HLA status, particularly HLA-B18, while patients with the HLA-B7 marker were normocomplementaemic (Trouillas *et al.*, 1976). These differences did not appear to be prognostically significant.

The myelinotoxic effects of sera from MS patients on CNS tissue cultures have been investigated particularly by Bornstein and his colleagues (Bornstein, 1973). Although demyelinating properties of sera correlate with disease activity, effects are not confined to MS. It has not been possible to identify consistent antibody or target antigens and 'serum myelinotoxicity' studied in this way now appears to be a non-specific phenomenon (Caspary, 1977). Effects of MS sera on neuronal polysynaptic function have also been described (Bornstein and Crain, 1965; Cerf and Carels, 1966). Initial hopes of developing a sensitive electrophysiological assay to investigate 'neuronal depressants' in MS were not born out, however, as subsequent studies clearly indicated the non-specificity for MS of these neurophysiological properties of serum (Crain, Bornstein and Lennon, 1975).

11.3 LIPID CHANGES

11.3.1 Brain

Lipid represents about 78% of the dry weight of human myelin (Smith, 1967). Awareness of the predominantly lipid nature of the myelin sheath excited early interest in the possibility that MS could be a disease of lipid of CNS myelin. Present data on the lipid composition of myelin have been summarized by Norton (1977a). The principal lipid classes present are cholesterol, phospholipid and galactolipid in a molar ratio of between 4:3:2 and 4:4:2. Cholesterol is the largest single lipid constituent of myelin and with galactolipids (cerebroside and sulphatide) and phospholipids (e.g. phosphoglycerides, plasmalogens, phosphosphingosides make up the myelin lipid profile. In order to interpret lipid changes in MS brain an understanding is required of the chemical changes found in myelin breakdown in different pathological situations. The chemical sequence of events when normal myelin is disrupted in both demyelinating and Wallerian-type reactions is remarkably uniform. The essential features are an initial phase of physical disruption of the sheath followed by a second phase of loss of and chemical degradation of myelin lipids, with the appearance of esterified cholesterol (Rossiter, 1955; Adams, 1969). A useful distinction is drawn between demyelination with breakdown of normal myelin and dysmyelination denoting breakdown of abnormally consistuted myelin (Poser, 1961). Although these patterns of myelin breakdown in brain and peripheral nerve are similar with early histophysical changes in both situations, cholesterol ester is detectable in

peripheral Wallerian degeneration by 6 days after nerve injury (see Hallpike, 1976) whereas breaking-down myelin in CNS retains normal staining characteristics with delay in the appearance of cholesterol ester for up to two months (Daniel and Strich, 1969). The finding of cholesterol ester in MS plaques (Cumings, 1953, 1955; Davison and Wajda, 1962) as well as in Schilder's disease (Suzuki, Tucker and Rorke, 1970) suggests strongly that demyelination in these conditions, as well as in postviral and postvaccinal reactions, affects chemically normal myelin. On the other hand, the dysmyelinating diseases (e.g. metachromatic leucodystrophy, Krabbe's disease, lipidoses) result from genetically determined inborn errors of metabolism and on this basis are clearly distinguishable from the acquired myelinopathies. Early ideas (Sperry and Waelsch, 1950) that loss of myelin in MS might represent an imbalance between formation and catabolism of myelin have largely been discounted on the basis of the overall stability of normal myelin membrane with isotopic evidence of an extremely low metabolic turnover of all the major myelin constituents in the mature animal (Adams and Davison, 1965). The biochemical hallmarks of active demyelination are the presence of cholesterol ester with concomitant decreases in free cholesterol, cerebroside, sulphatide, phospholipids and myelin-specific G_7 ganglioside (Yu, Ledeen and Eng, 1974). The cholesterol ester is accumulated within phagocytic cells which persist in plaques although, finally, old lesions become acellular with a complete loss of all reactivity associated with myelin lipid (see Chapter 8 by C. W. M. Adams for detailed account). Support for a generalized abnormality of myelin lipid in MS has been sought particularly through analysis of macroscopically normal white matter. Minor changes, e.g. reduced plasmalogens (Yanagihara and Cumings, 1969), have been reported but, in general, qualitative changes have not been found and reduced yields of some lipid moieties are best explained by the inclusion of undetected discrete lesions (Norton, 1977b). Myelin isolated from normal-appearing white matter of MS brain contained normal amounts of individual lipids (Suzuki *et al.*, 1973; Fewster, Hirano and Mead, 1976). Nevertheless, data derived from the study of myelin fractions are not conclusive as the preparative procedures may be weighted in favour of residual normal tissue (see Gregson, Chapter 1).

11.3.2 CSF

Cerebrospinal fluid lipids have been studied in MS on the grounds that disease affecting CNS myelin might produce changes of diagnostic or prognostic significance in the CSF lipid profile. These data have largely been reviewed by Tourtellotte (1970). Total lipids were increased in the CSF in clinically definite MS, such changes being most evident for free cholesterol, cerebrosides, sphingomyelin and cephalins (Tourtellotte and Haerer, 1969). Similar findings were reported by Clausen and Fog (1969) who also showed

that the cerebroside:lecithin ratio in CSF was increased in MS compared with normal subjects and other neurological diseases. Pedersen (1974) found a small increase in cholesterol ester in CSF in severer MS. However, this finding was not of value in individual case assessment and appeared to be an indirect parameter of disease activity.

11.3.3 Blood

Most studies indicate that serum cholesterol in MS is normal (Zilkha, 1970). Early reports of plasma phospholipids in MS were conflicting and more recent evidence points strongly towards normal phospholipid profiles (Tichý, Vymazal and Michalee, 1969; Karlsson, Alling and Svennerholm, 1971). Serum levels of cerebroside, a 'myelin-typical' galactolipid, have not been found to be increased in MS (Rathke and Jones, 1974). Attempts to date to study disease activity by serum measurement of putative markers of CNS lipid breakdown have been unsuccessful. The position with regard to fatty acids in myelin membrane and in serum in MS is considered below.

11.4 FATTY ACIDS

Altered fatty acid (FA) composition of white matter lipids, with reduced proportions of unsaturated FAs in phospholipid fractions, has been reported in MS (Gerstl *et al.*, 1961; Baker, Thompson and Zilkha, 1963). It was also suggested (Gerstl *et al.*, 1970; Yatsu and Moss, 1970) that defective FA elongation, possibly during myelination, could reduce the stability of myelin membrane and predispose to multiple sclerosis. Other findings (Alling, Vanier and Svennerholm, 1971; Suzuki *et al.*, 1973) have not confirmed any consistent pattern of FA abnormality and it is likely that these minor lipid changes in normal-appearing white matter which have been described reflect microscopic lesions and are not indicative of any generalized underlying biochemical abnormality.

A reduced percentage of linoleate (C18:2) has also been found in plasma lipids in MS patients, the degree of reduction in linoleate appearing to correlate with activity of the disease process (Baker *et al.*, 1964; Belin *et al.*, 1971). Studies of FA metabolism in MS, reviewed by Thompson (1972) and Smith and Thompson (1977), led to the proposition that there may be an inborn error of handling of unsaturated FAs in MS resulting in cell membrane abnormalities predisposing to multiple sclerosis. Rats rendered deficient in polyunsaturated FAs have been shown to have increased susceptibility to experimental allergic encephalomyelitis (Clausen and Moller, 1967). Karlsson *et al.* (1971) were unable to find any deficiency of polyunsaturated fatty acids (PUFA) in MS plasma lipids. Reduced serum linoleate levels have also been reported, however, in patients with non-neurological diseases (Love *et al.*, 1974). Extensive clinical trials of dietary linoleate supplementation in MS (Millar *et al.*, 1973; Bates *et al.*, 1977, 1978) have been consistent in suggesting

a marginal reduction in duration and severity of relapses but with no effect of such treatment on overall disability or progression of the disease. Polyunsaturated fatty acids might act in a number of ways in MS. Restitution of PUFA deficiency could have some stabilizing effect on the oligodendrocyte or myelin membrane. PUFAs have been shown to have immunoregulatory effects in experimental allergic encephalomyelitis (EAE) and other models of cell mediated immunity (Mertin and Meade, 1977). Linoleic acid is a precursor of prostaglandin and supplementation may promote formation of prostacyclin, a natural inhibitor of platelet aggregation and a vasodilator (see Hallpike, 1980). Linoleic acid has been found to affect the electrophoretic mobility of lymphocytes and red cells of subjects with MS in a manner which has been claimed by Field and his colleagues (Field, Shenton and Joyce, 1974; Field, 1977 to distinguish MS from other neurological diseases. The underlying hypothesis of a FA deficiency in MS, the role of PUFA in MS therapy and diagnostic tests for MS involving FAs are very controversial and the subject of conflicting reports.

Swank (1950) drew attention to a possible relationship between the prevalence of MS and the consumption of animal fats. Diets rich in animal fats also tend to be deficient in PUFAs (Sinclair, 1956). Employment, in MS, of a PUFA-enriched low animal fat diet was reported to be associated with fewer relapses and a lower death rate in an uncontrolled personally treated series of patients (Swank, 1970).

11.5 PLATELET STICKINESS

Although the notion (Putnam, 1935, 1937; Fog *et al.*, 1955) that venular thrombosis might be a pathogenetic factor in MS plaque formation has been firmly discarded, the usual occurrence of centrally placed venules within plaques (see Adams, 1972; also Chapter 8) as well as increased platelet stickiness have served to maintain some interest in vascular phenomena in MS. The demonstration in MS of abnormal platelet adhesiveness *in vitro* (Nathanson and Savitsky, 1952; Caspary *et al.*, 1965; Wright, Thompson and Zilkha, 1965; Millar, Merrett and Dalby, 1966) led to speculation that platelet changes *in vivo* might cause venular occlusion and contribute to plaque formation. Nevertheless, patients with MS do not show other clinical features of increased coagulability. Studies of blood coagulation and fibrinolytic activity, in fact, showed slightly reduced clotting and increased fibrinolysis in MS in relapse (Feldman, Izak and Nelken, 1957; Menon, Dewar and Newell, 1969). A trial of Atromid in MS effectively reduced serum cholesterol and total esterified FAs but had no effect on platelet stickiness *in vitro* or the course of the disease (Prineas *et al.*, 1967). Hence, there is no consistent evidence that these platelet and coagulation phenomena have any primary role in MS. Nevertheless, Thompson and his colleagues (Thomson, 1975 have drawn attention to a sharp inverse relationship between platelet stickiness and serum

linoleate levels in patients with MS. The biosynthesis of prostaglandins by platelets from patients with MS has been shown to be reduced, without alteration of the important ratio PGE_1/PGE_2 or of evidence of abnormal platelet aggregation (Srivastava, Fog and Clausen, 1975). Although such data indicate a possible role for FAs in the aetiopathogenesis of MS, as suggested by Thompson, strong reservations remain about the specificity for MS and causal nature of the findings.

11.6 ENZYME CHANGES

Early studies of neuroprotease activity demonstrated the presence in brain of intracellular proteinases, cathepsins with endopeptidase activity. A brain cathepsin with an acid pH optimum (Ansell and Richter, 1954) is associated with the lysosomal fraction (Marks and Lajtha, 1963) and closely resembles cathepsin D, an enzyme of lysosomal origin isolated from other organs (De Duve, Wattiaux and Baudhuin, 1962). Lysosomes are widely distributed in brain, being readily displayed in normal and pathological neurones, in astrocytes and oligodendrocytes (Koenig, 1969). Increased acid proteinase activity has been demonstrated in 'active' MS plaques (Hallpike and Adams, 1969; Einstein *et al.*, 1970). It appears that acid proteinase may be instrumental in degrading MBP and contributing to myelin breakdown in the evolving MS lesion (Einstein *et al.*, 1972; Hallpike, 1972). Most of the proteolytic activity is closely associated with acid phosphatase and largely reflects local lysosomal changes in macrophages. Neutral proteinases, present in myelin (Riekkinen and Clausen, 1969) are mainly unaltered in MS plaque tissue. Changes in cerebral lysosomal activity have also been described in apparently normal white matter in MS, i.e. lysosomal acid proteinase, β-glucuronidase and arylsulphatase (Cuzner and Davison, 1973) and β-glucosaminidase (Allen and McKeown, 1979). Reports, however, of more widespread loss of MBP, concomitant with these increases in hydrolase activity, have been conflicting (see Norton, 1977b). Nevertheless, indications of alterations to the MBP in macroscopically normal white matter have been obtained (Althaus, Pilz and Müller, 1973; Cuzner *et al.*, 1976) with a suggestion that such changes could be associated with astrocytosis. Greatly increased activity of a thiol-dependent carboxypeptidase in and around MS plaques was considered to be of astrocytic origin (Hirsch and Parks, 1979). Elevation of cerebral proteinase activity also occurs in experimental allergic encephalomyelitis (Kerekes, Feszt and Kovacs, 1965; Gabrielescu, 1969) and is associated with selective loss of MBP in that condition (Rauch, Einstein and Csejtey, 1973). Acid proteinase activity in CSF was found to be increased in acute and chronic MS, as well as in patients with seizure disorders and stroke, while raised neutral proteinase activity in CSF was confined to acute MS (Rinne and Riekkinen, 1968). There have been other reports of increased neutral proteinase activity in the CSF in MS (Cuzner, Davison and Rudge,

1978). Other enzymes of lysosomal origin, glucosaminidase, β-galactosidase and α-mannosidase, were not consistently altered in CSF, although patients with one or two recent relapses or with later onset of MS had the highest activities (Hultberg and Olsson, 1979). The proteinase inhibitor, alpha$_1$-antitrypsin, was found to be decreased in MS cerebrospinal fluid although serum levels of the inhibitor were normal (Price and Cuzner, 1979). Increased neutral proteinase activity in blood leucocytes occurs in acute MS and is also found in other neurological diseases characterized by rapid destruction of neural tissue Cuzner *et al.*, 1975; Czernicki *et al.*, 1979). Acid phosphatase and β-glucuronidase were found to be increased respectively in the granulocytes and lymphocytes of patients with MS in remission (Riekkinen, Palo and Asikainen, 1977). The significance of these leucocyte changes in MS is uncertain but may be linked to the removal of CNS antigen from the systemic circulation. The enzyme 2',3'-cyclic nucleotide 3'-phosphohydrolase (2',3'cAMPase) is localized in the myelin sheath (Kurihara and Tsukada, 1967) and provides a useful biochemical marker for myelin. Reduction in 2',3'cAMPase activity has been reported in multiple sclerosis myelin (Riekkinen *et al.*, 1972). This enzyme has been detected in normal CSF and, although increased activity occurs in MS, correlation with disease activity appears to be poor (Sprinkle and McKhann, 1978; Banik, Maudlin and Hogan, 1979 . The finding that erythrocytes from a group of patients with MS showed decreased activity of glutathione peroxidase, a selenium-dependent enzyme involved in membrane stability, has led to a suggestion that topographic differences in the availability of selenium may be relevant to MS (Shukla, Jensen and Clausen, 1977).

11.7 MYELIN BASIC PROTEIN IN CSF

Myelin basic protein and MBP fragments have been found in CSF in acute multiple sclerosis using radioimmunoassay (RIA) methods (McPherson, Gilpin and Seland, 1972; Cohen *et al.*, 1976; Whittaker, 1977; Cohen *et al.*, 1978). Measurement of MBP in CSF appears to correlate well with exacerbations and progression of the disease and may be a useful indicator of disease activity and effects of therapy (Brooks *et al.*, 1979). Myelin basic protein has also been measured in serum of patients after cerebrovascular accidents, the levels being related to severity and prognosis (Palfreyman *et al.*, 1979). This study also provided evidence of the presence, in patients with a past history of stroke, of serum MBP-binding activity consistent with the presence of antibodies to basic protein. Electron-microscopic examination of ultracentrifugal sediments of CSF may reveal cellular elements of neural origin, including myelin fragments, in multiple sclerosis (Herndon and Johnson, 1970). Although impractical for routine diagnostic use, such findings point to possible sources of MBP and other brain-specific antigens in the CSF.

11.8 IMMUNE COMPLEXES AND T CELL RATIOS

Immune complexes (IC), characteristically composed of immunoglobulin and complement, are known to be present in the circulation and to be deposited on the basement membrane of capillaries or larger vessels and to be implicated in the pathogenesis of an increasing number of diseases involving the immune system. Study of IC may permit identification of antigen related to the cause of the disease although IC frequently contain autoantigen in the form of self-associated IgG. Delayed clearance of IC may reflect immunodeficiency in terms of macrophage function while the presence of IC may also inhibit the development of an immune response (Masson, 1978). Immune complexes have been found in brain (Tavolato, 1975), in sera (Tachovsky *et al.*, 1976) and in CSF (Deicher *et al.*, 1979; Coyle *et al.*, 1980) of MS patients. Circulating deposits of IgG-C'3 were found in roughly 45% of sera from patients with MS, optic neuritis and Guillain-Barré syndrome, compared with 15% of normal sera, with little correlation, however, between the presence of IC and clinical status in the MS cases (Tachovsky *et al.*, 1976). Immune complexes have also been detected in CSF in approximately 50% of MS patients during exacerbations, there being a concomitant decrease of a subpopulation of IgG Fc receptor-positive lymphocytes (F_c cells) thought to have immunosuppressor potential (Coyle *et al.*, 1980). Reduced levels of CSF complement, particularly C'2,C'3, occurring in MS (Bammer, 1966) may be an indication of IC formation.

Recent work suggests that the balance of subpopulations of T lymphocytes is important in maintaining the normal function of the human immune system. Changes have been demonstrated in these subpopulations in various diseases by means of Fcγ and Fcμ receptor markers as well as by the use of monoclonal antibodies. The recent availability of such antibodies, i.e. of the OKT series, produced by hybridization/cloning techniques, is now being exploited in many clinical immunological laboratories in the study of pan T (OKT_3), helper/inducer (OKT_4) and suppressor (OKT_8) cell values and ratios in diseases involving possible immune dysregulation. In MS, a population of suppressor T cells, detected in blood by OKT monoclonal antibody, was found to be selectively depressed in 11/15 patients with clinically 'active' disease and in only 1/18 patients in an 'inactive' phase (Reinherz *et al.*, 1980). It has therefore been proposed that changes in population of suppressor T lymphocytes may contribute to the pathogenesis of the disease.*

11.9 AMINO ACIDS

There does not appear to be any relationship between MS and genetically determined abnormalities of amino acid metabolism. Nevertheless, amino acid concentrations within the brain-CSF compartment are particularly

* See also Chapter 12 by S. Leibowitz for full discussion of T cell studies in MS—Ed.

sensitive to blood–brain barrier alterations accompanying structural CNS disease (Rapoport, 1976) and CSF amines in MS have been extensively studied (Lumsden, 1972). Minor general reductions in CSF amino acids were reported for some MS cases (Williams and Matthews, 1965). The CSF protein serine residue appears to correlate closely with the CSF IgG and it has been suggested that the serum/CSF protein serine ratio could be developed as an index of disease activity (Poser *et al.*, 1975). A relatively selective reduction in CSF tryptophan has been reported in CSF in MS and, to a lesser extent, in motor neurone disease, leading to the suggestion that 5-hydroxytryptamine (5-HT) could be depleted in MS brain (Monaco *et al.*, 1979). Of possible relevance in this context is the evidence that the encephalitogenic tryptophan-containing amino acid sequence of MBP, the 'tryptophan peptide', closely resembles a CNS receptor site for 5-HT and the proposition arising from this that some symptoms in MS may be due to immunopharmacological block of 5-HT receptor sites (Lennon and Carnegie, 1971; Carnegie and Mackay, 1975).

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CHAPTER TWELVE

The Immunology of Multiple Sclerosis

S. Leibowitz

12.1 Introduction
12.2 Immune function in MS
12.3 Autoimmuninity in MS
12.4 The role of immunity in pathogenesis
12.5 General conclusions

12.1 INTRODUCTION

Multiple sclerosis is an inflammatory disease in which immunoglobulin is synthesized within the nervous system. This activity is a central feature of the immunopathology which it shares with many persistent infections, including syphilis and subacute sclerosing panencephalitis (SSPE). It has been calculated that 30 mg day^{-1} of IgG is produced intrathecally in the average patient and values as high as 207 mg day^{-1} have been reported (Tourtellotte and Ma, 1978; Ewan and Lachmann, 1979). However, all attempts to define the antigen have been unsuccessful and it is still not clear whether we are dealing with a response to a virus, an autoimmune reaction or some other manifestation of hypersensitivity.

Despite failure convincingly to demonstrate any infectious agent (see ter Meulen, Chapter 9), there is still a strong presumption that MS is a viral disease and, if so, the natural history of MS will at least partly be determined by immunological factors. This relates to such well-known features of MS as the presumed long latent period, the chronicity of the disease and the characteristic clinical pattern of exacerbation and remission. There is the geographical distribution of MS (see Kurtzke, Chapter 3), indicating that populations in different parts of the world vary very greatly in their susceptibility to the disease (viz. the tropics vs. temperate zones). Finally, there is also the association between histocompatibility phenotype and susceptibility which has yet to be satisfactorily explained (see Stewart and Kirk, Chapter 4). All these—and many other aspects of the disorder—have immunological implications and justify the emphasis currently placed on immunity in MS research.

A second and equally powerful stimulus is provided by the notion that the demyelinating process is not due to direct injury, but, rather, is mediated by

some form of hypersensitivity. Although there is little direct evidence to support this view the hypothesis has proved remarkably durable and survived more than five decades of disappointing endeavour. Historically, it arose from two separate lines of investigation. The first was the study of the meningoencephalitis which occasionally follows acute viral infections or vaccination. This is associated with perivenous demyelination and, since the same end result is triggered by many different viruses, it is generally assumed that the final common path must be immunological. Similar reasoning may be invoked to explain the demyelination in chronic viral infections and, although the lesion of post-infectious encephalomyelitis is quite unlike that of MS, the argument has a certain plausibility. However, there is remarkably little evidence bearing on this matter one way or the other, and it is difficult to see how the question can be resolved while we remain uncertain whether or not the disease is due to an infectious agent.

The second analogy is with experimental allergic encephalomyelitis (EAE), a condition in which the demyelinating lesion is produced in the CNS by sensitization to myelin basic protein (MBP). The resemblance between acute EAE and MS is not particularly striking, but a chronic form of the disease has recently been described which produces large areas of demyelination and provides a more convincing parallel with the MS plaque (Raine, Chapter 13). This has rekindled interest in the autoimmune hypothesis and encouraged those who regard EAE as a model of the human disease.

It will be seen that the immunology of MS covers a wide field and is concerned with four main problems:

(1 Viral immunity and the search for an infectious agent.
(2 The nature and significance of the immune response in the CNS.
(3 Immune function in the MS patient and its relation to the natural history of the disease.
(4 The role of autoimmunity and hypersensitivity in the genesis of the MS plaque.

Viruses and the immune response in the CNS have been considered in some detail in previous chapters (Chapters 9 and 10) and in this section we shall confine our attention to two questions. Firstly, whether immune function in MS is disturbed—the nature of the disturbance and how it relates to the clinical pattern of the disease; and secondly, whether autoimmunity can explain the demyelinating process or any other aspect of the pathology.

12.2 IMMUNE FUNCTION IN MS

12.2.1 Lymphocyte sub-populations in the peripheral blood

Lymphocytes perform a variety of functions and constitute a remarkably

heterogeneous population of cells despite their uniform appearance. In recent years considerable progress has been made in identifying the different subgroups using surface markers and a battery of *in vitro* tests. The main division is into B or bone-marrow-derived cells and T-cells derived from the thymus and each is made up of a number of functionally distinct subsets.

In MS the total lymphocyte count in the peripheral blood is normal or slightly reduced and the level is unrelated to disease activity. The proportion of T cells is essentially unaltered, although some reports suggest that it is depressed during exacerbations. This may be related to the fact that they appear in increased numbers in the CSF. The B cell population is also normal or reciprocally increased (Lisak *et al.*, 1975a; Oger *et al.*, 1975; Sandberg-Wolheim and Turesson, 1975; Reddy and Goh, 1976; Naess and Nyland, 1978; Symington *et al.*, 1978; Weiner and Schocket, 1979; Kateley and Bazzell, 1979; Mar *et al.*, 1979; Sagar and Allonby, 1979).

In addition, there is evidence that the relative proportions of the T cell subsets may be altered, particularly in the active phase of the disease. The first change to be described was a reduction in the number of 'active' or 'avid' rosetting cells (T_A). These have a high affinity for sheep erythrocytes and form rosettes more rapidly than other T lymphocytes. T_A cells can be identified by incubating peripheral blood lymphocytes with sheep red cells for 5 minutes, instead of the usual 18 hours and enumerating the 'early' rosettes. They are thought to represent a subset of lymphocytes actively engaged in cell-mediated responses and more directly involved than the T-cell population as a whole (Wybran and Fudenberg, 1973). The proportion of these cells in the circulating blood is reduced in MS and this is particularly marked in the acute phase of the disease (Oger *et al.*, 1975; Antel *et al.*, 1978a; Kateley and Bazzell, 1979; Traugott, Scheinberg and Raine, 1979). This suggests a depression of cell-mediated immunity associated with disease activity, although the significance of the finding is somewhat reduced by the fact that many investigators have observed a similar depression in neurological controls. The general impression is that the effect is secondary and related to CNS tissue damage, viral infection or a combination of both (Goust *et al.*, 1978; Offner *et al.*, 1978).

12.2.2 Suppressor cells

The most interesting finding in the peripheral blood is the observation that the number of suppressor cells fluctuates during the course of the illness. The function of this subset (T_S), is to modulate the immune response, and together with the helper T cells (T_H) they form part of a complex regulatory system that controls antibody production. Helpers as well as suppressors are produced following immunization, and these two subsets contribute towards

About 75% of peripheral blood lymphocytes have Fc for receptors for IgG and 10–15% for IgM. Suppressor cells are found in the Tγ fraction and they can be identified by the fact that they bind aggregated IgG or IgG immune complexes. In practice they are usually assayed by exploiting their capacity to form rosettes with bovine red blood cells coated with a rabbit antibody. Increased levels of T cells have been reported in MS with this method, but the increase apparently bears no relation to the phase of the disease (Santoli *et al.*, 1978; Merill *et al.*, 1980). However, when followed serially, patients showed a reduction of cells in the acute attack, followed by an increase as they went into remission. A similar pattern has been described in the CSF (Huddlestone and Oldstone, 1979).

It is possible to elaborate a number of more or less plausible hypotheses linking Ts levels and disease activity in MS. There is normally a balance between helper and suppressor cells in the circulation, and one might envisage a situation in which the equilibrium is disturbed, releasing potentially autoreactive clones and triggering an immune response. A mechanism of this kind has been observed in autoimmune disease in experimental animals. For example, the onset of haemolytic anaemia in NZB/NZW mice is preceded by a loss of suppressor activity and specifically recruited suppressor cells will inhibit the passive transfer of EAE in rats (Bernard, 1977). There is also evidence that autoimmunity in some human diseases, such as systemic lupus erythematosus, may be linked to a decline in Ts activity.

Most viral infections appear to increase suppressor activity and this has been observed with the Epstein-Barr virus in mononucleosis. However, it is conceivable that some infections may depress Ts levels. Arnason observed a herpes type II infection in one of his MS patients and found that recurrence of the virus infection was associated with a flare-up of the disease. Each herpetic eruption was followed within a few days by recrudescence of neurological signs. There were three such cycles and on the third occasion suppressor cell activity was measured and found to be absent. This decline coincided with reactivation of acute MS, suggesting that the attacks were precipitated by virus-induced depression of the suppressor mechanism (Antel *et al.*, 1978a). A similar phenomenon has been observed in animals with EAE. If rats are immunized with herpes virus followed by the injection of an emulsion of spinal cord in Freund's adjuvant they develop EAE normally. However, re-injecton of the virus after recovery may lead to a recrudescence of the disease. From this, it appears that viral infection may induce a decline in suppressor activity, reactivating the autoimmune response and producing a second bout of EAE (Hochberg, Lehrich and Arnason, 1977).

The observations in MS, referred to above, are of great interest but previous experience in this field should make one cautious and these findings require further confirmation. Apart from the technical difficulty of measuring suppressor activity there is the clinical problem of distinguishing between

measles infection. Similar results with the migration inhibition tests have been obtained by many, although not all, subsequent investigators (Ciongoli, 1977; Platz, 1977; Lisak *et al.*, 1978; Fucillio *et al.*, 1978; Myers, Ellison and Holevoet, 1978; Visscher *et al.*, 1979). Lymphocyte transformation is more difficult to assess, since normal cells are only weakly stimulated by this virus *in vitro* and the results in MS have been equivocal (Dau and Peterson, 1970; Knowles and Saunders, 1970; Cunningham-Rundles *et al.*, 1977; Symington and Mackay, 1978; McFarland and McFarlin, 1979). Further evidence of an impaired cellular response to measles comes from the study of T cell cytotoxicity. When cells in culture are infected with measles they express viral antigens on their surface and may be damaged by peripheral blood leukocytes from immunized individuals. Three different kinds of cytotoxicity can be demonstrated—antibody-mediated K-cell killing and damage due to monocytes or specifically sensitized T cells. The first two are unaffected in MS, but T cell cytotoxicity is significantly impaired (Ewan and Lachmann, 1977; Huddlestone and Oldstone, 1978).

Although measles has attracted most attention, cell-mediated responses to other viruses may be reduced as well. All the common viruses have been studied and there is evidence of depressed CMI to mumps, parainfluenza and vaccinia (Cunningham-Rundles, Dupont and Posner, 1975; Ciongoli, 1977; Walker and Cook, 1978, 1979; McFarland and McFarlin, 1979; Platz, 1977). However, there is disagreement on this point and some maintain that measles is a special case. The most reasonable view would seem to be that the depression of cell-mediated immunity (CMI) is marginal, with some viruses more affected than others. Once again it appears to be a secondary phenomenon, since it is not present early in the disease, while in long-standing cases it is related to the degree of disability. Moreover, a similar depression of CMI has been observed in neurological controls (Myers *et al.*, 1978; Symington and Mackay, 1978; McFarlane and McFarlin, 1979; Walker and Cook, 1978, 1979).

(d) One additional aspect of lymphocyte function in relation to viruses may be mentioned at this point, although its significance is still uncertain. Levy, Auerbach and Hayes (1976) found that peripheral blood lymphocytes from MS patients adhered to measles-infected epithelial cells in greater numbers than normal. There was no overlap between MS and control values, suggesting that the lymphocyte adherence determination (LAD) test might prove valuable in diagnosis. This phenomenon has been confirmed by others, although their results have not been as clear-cut and there has been at least one negative report (Offner *et al.*, 1977; Daly and Desai, 1978; Kinman and Link, 1979; Dore-Duffy *et al.*, 1979). There is normally a tendency for peripheral blood leukocytes to adhere to measles-infected cells and it is not clear what this increased stickiness signifies. It is also uncertain whether the adhering cell is a monocyte or lymphocyte. The phenomenon is completely unexplained and its main interest lies in the question of whether it can be refined into a useful clinical test.

infected with rubella. However, it is not an immunoglobulin and the effect cannot be produced with normal serum containing antimeasles antibody.

Not everyone is convinced of the existence of these elusive factors and Walker and Cook (1979) deny that inhibitory phenomena are more frequent with MS than normal sera. The nature of these blocking effects is far from clear and it may be that they are at least partly due to immune complexes, which are known to inhibit cellular responses. Their significance *in vivo* is uncertain, although one may speculate on the possibility that they contribute to the depression of CMI. It is also possible that some of the earlier reports of diminished responses *in vitro* may have been due to the use of autologous serum.

12.2.6 Lymphocytotoxic antibody

Many MS sera contain an autoantibody capable of reacting with human lymphocytes. It is a cold-reactive IgM which binds optimally at 15°C and can be demonstrated by the micromethod used in tissue typing. Serum and peripheral blood lymphocytes are incubated in microtitre plates, complement is added and the number of dead cells determined by their uptake of a vital dye. The incidence of lymphocytotoxic antibody (LCA) in MS is about 40% compared with 5% in the normal population, although figures as high as 67% have been reported (Kuwert and Bertrams, 1972; Schocket *et al.*, 1977). The significance of this antibody is unknown, but one line of evidence suggests that it may be related to infection. Schocket and Weiner (1978) found a high incidence in the households of patients with MS and noted that there was no difference between consanguinous and non-consanguinous individuals living under the same roof. On the other hand, the figure for sibs living elsewhere was reduced, suggesting that the antibody was a response to a transmissable agent or some other environmental factor.

Lymphocytotoxic antibodies occur in systemic lupus erythematosus, rheumatoid arthritis, pernicious anaemia and several other diseases of obscure aetiology. They are also present in patients with acute viral infections and after immunization with viral vaccines (Kreisler, Hirata and Terasaki, 1970). In neurological disease apart from MS, an increased incidence has been noted in myasthenia gravis and SSPE. The relation of these antibodies to immunity has been closely studied in systemic lupus erythematosus, where it is present in nearly all patients. The evidence suggests that LCA is an autoantibody induced by viral infection of lymphoid cells, and some observers have noted a correlation between antibody and disease activity. However, it now appears that the lymphopaenia and depressed CMI in this disease are not due to IgM, but to a similar lymphocytotoxic IgG antibody which reacts at body temperature. The changes in the lymphocyte subpopulations in MS cannot be attributed to cytotoxic antibody, since it only

components and their presence in the sheath has been confirmed by immunohistochemical methods. The glycolipids are also immunologically active, although they are incomplete antigens and will only elicit antibody when injected together with a protein carrier. The three main haptens are galactocerebroside, sulphatide and ganglioside (GM1 and GM4). Specific antisera can be raised to each of these components in rabbits, the activity being mainly directed to the oligosaccharide part of the molecule.

Since these compounds are relatively insoluble in aqueous media, the most convenient method for detecting antibody is complement fixation, although it is also possible to incorporate the haptens into liposomes and use the technique of liposome lysis (Kinsky, 1972). Galactocerebroside is present in highest concentration and is the characteristic lipid of the myelin sheath. It is a powerful hapten and the complement fixing anti-brain antibody obtained by immunizing an animal with whole brain or particulate myelin is largely directed against this compound.

There are, therefore, a wide range of antigenic substances in myelin, many of them potential autoantigens. However, most investigators have until recently limited themselves to the study of the encephalitogenic basic protein.

12.3.2 Sensitization to basic protein

It has generally been assumed that, if MS is an autoimmune disease, the most likely antigen is MBP. Ever since the purified encephalitogen became available attempts have been made to demonstrate antibody to it in the serum. A variety of methods has been used including passive haemagglutination, the Farr technique, co-precipitaton, immune adherence, radioimmunoelectrophoresis and radioimmunoassay. In general, the results have been disappointing and largely negative (McPherson and Carnegie, 1968; Lisak *et al.*, 1978; Biggins, Taylor and Caspary, 1978). Where positive results were obtained, they were accompanied by similar findings in neurological controls, suggesting that the antibody was secondary to CNS damage (Field, Caspary and Ball, 1963; Caspary and Chambers, 1970). The failure to demonstrate circulating antibody with radioimmunoassay—the most sensitive and direct method—is particularly significant.

On the other hand, positive findings have been reported using indirect and less well-established procedures. Sheremata *et al.* (1978b) claimed that antibody could be detected by double diffusion in agar if the immunoglobulin was first separated by electrophoresis on polyacrylamide. They explained the failure of whole serum to give this reaction by the presence of myelin degradation products, which combined with the antibody and inhibited combination with the test antigen. Antibody was only found in the convalescent phase of the disease.

A curious inhibition test has also been described by McPherson, Libird and Seland (1975) who observed that binding of I-labelled MBP to normal sheep

lymphocytes was inhibited by concentrated serum from patients with MS. In longitudinal studies on several patients they found that inhibitory activity was associated with clinical improvement and that serum from patients in remission was more inhibitory than from patients in relapse. The nature of this unusual blocking activity is unknown, although it would seem unlikely that it is due to specific antibody.

Since immunoglobulin is synthesized intrathecally in MS it would be logical to look for antibody in the CSF rather than in the serum. However, this is a matter of some difficulty, since the protein concentration in CSF is low and it is necessary to concentrate the fluid before testing. Attempts to measure anti-MBP antibody in the CSF by radioimmunoassay have given conflicting results (Panitch, Hafler and Johnson, 1978; Gutstein and Cohen, 1978).

Finally, there is the recent report of Frick and Stickl (1980) of antibody to MBP detected by antibody-dependent lymphocyte cytotoxicity. In this test chromium-labelled chicken erthrocytes are coated with the antigen and exposed to the patient's serum; mouse or human lymphocytes are added and cytotoxicity measured by chromium release. Positive findings were obtained in nearly 80% of all cases of MS, the figure rising to 94% in those with active disease. Positive results were also reported with CSF. Antibody was not detected in the serum of normal subjects and was only present in 8% of neurological controls. On the other hand, nearly a third of all patients with syphilis (without neurological involvement) gave positive results. However, these remarkable findings require confirmation. The K cell assay is sensitive but, as with all cellular tests, it is capricious and it is difficult to see why it should be successful in detecting antibody not found by radioimmunoassay. On the other hand, if the findings are correct the K cell cytotoxicity may prove a valuable diagnostic test, and the entire question of antibody to MBP in MS will have to be reconsidered.

12.3.3 Cellular immunity to myelin basic protein

There has been a natural reluctance to skin test patients with the encephalitogenic protein because of the potential hazard, but the evidence —as far as it goes—does not suggest that there is hypersensitivity to this antigen in MS. CMI has been studied with a variety of *in vitro* tests, including lymphocyte transformation and migration inhibition assays (Hughes, Caspary and Field, 1968; Dau and Peterson, 1970; Bartfield and Atoynatan, 1970; Stangaard and Jorgensen, 1972; Uyeda and Murphy, 1976; Lisak *et al.*, 1978; Clanet *et al.*, 1979; Hughes *et al.*, 1979). No concensus has emerged and, although most workers have reported negative findings, others—notably Sheremata and his group—have obtained positive results with the macrophage migration inhibition test and claim that it correlates with disease activity. Similar, if less impressive, results were obtained with lymphocyte transformation (Sheremata, Cosgrove and Eylar, 1974; Colby *et al.*, 1976;

Sheremata *et al.*, 1978a, b). To compound the confusion, a two-stage procedure has recently been introduced in which stimulation of ConA is measured after pretreatment with MBP. Using this method, Wicher, Olzewski and Milgrom (1979) found that blastogenesis was enhanced in active disease and depressed in remission.

Several other indicators of CMI have been tried, including the specific generation of 'active' T cells. In this test the number of 'early' rosetting cells is counted before and after the addition of antigen to peripheral blood lymphocytes and an increase is taken to indicate specific sensitization. Evidence of hypersensitivity to MBP has been obtained in MS and in patients with CNS tumours or brain damage due to cerebrovascular disease (Hashim *et al.*, 1978; Offner *et al.*, 1978; Kateley and Bazzell, 1979).

For many years Field has maintained that cellular immunity to MBP can be demonstrated by means of a macrophage electrophoretic migration assay (MEM). The patient's lymphocytes are exposed to antigen and the supernatant added to a suspension of guinea-pig macrophages. The rate of migration of these cells in an electric field is then measured in a cytoelectrophorometer and sensitization results in slowing (Field and Caspary, 1970). There is nothing inherently improbable in this assay, since lymphocytes are known to release lymphokine factors affecting macrophages and there is no reason why this should not affect their surface charge. However, cytoelectrophoretic measurements on macrophages are technically difficult and unreliable and many laboratories have been unable to repeat this work. On the other hand confirmation has been obtained from at least two centres (Meyer-Rienecker, Jenssen and Werner, 1979). The range of responders is wide and positive results have been obtained in many neurological diseases, and in patients with neoplasms and with acute or chronic infections.

Cells capable of binding MBP can also be detected by means of a rosetting test with antigen-coated erythrocytes. They are increased in MS during the acute attack, for some months afterwards and in chronic progressive disease. The nature of the antigen-binding cell is not known and it is uncertain whether this test reflects cell-mediated immunity, a B cell response or fluctuations in the surface properties of lymphocytes (Vandenbark *et al.*, 1979).

It is difficult to know what to make of the data on MBP. Antibody cannot be detected by radioimmunoassay (although the K cell assay is positive) and the evidence on cell-mediated immunity is conflicting. However, even the positive findings offer only limited support for the idea that MS is the human equivalent of EAE, since a similar degree of sensitization is often observed in neurological controls. All the indications are that an immune response to MBP, if it occurs at all, is the result and not the cause of the lesion in the CNS.

12.3.4 Glycolipid haptens

The earliest attempts to demonstrate anti-brain antibody in MS were made using complement fixation and alcoholic extracts of CNS tissue. These studies yielded little beyond suggesting that antibody to glycolipid might be present in some neurological diseases (Lumsden, 1972). Interest in these compounds has revived recently with the demonstration that antibody to galactocerebroside will demyelinate cultures of neural tissue (Dubois-Dalque, Niedieck and Buyse, 1970 and that prolonged immunization produces a demyelinating neuropathy in rabbits (Saida *et al.*, 1979). Although purified myelin glycolipids are readily available, there has been little systemic investigation of autoantibody in neurological disease. These compounds are difficult to investigate, since they are relatively insoluble in aqueous media and have a small molecular weight. This means that the usual stock-in-trade of the immunologist—diffusion in agar, precipitation and the like—are only of limited use. To produce a reaction the hapten is mixed with other lipids (generally cholesterol and lecithin) to form a micellar suspension. They facilitate antibody binding and secondary reactions such as complement fixation, but multiply the number of variables and complicate experimental design (Alving, 1977). Antibody can also be measured by incorporating the hapten into liposomes containing a marker trapped in the internal aqueous phase. In the presence of complement and specific antibody, these vesicles undergo lysis and the marker is released (Kinsky, 1972). This is an elegant method capable of fine discrimination, but there is no evidence that it is more sensitive than complement fixation.

There have been several reports of antibody to glycolipid in MS. Dupouey, Schuller and Coulon-Morelec (1972) found antibody to galactodiglyceride, cross-reacting with galactocerebroside in 16% of patients and 8% of controls; and similar figures have been reported for ganglioside using passive haemagglutination (Yokoyama, Trams and Brady, 1962). Nearly 60% of patients' sera react with digalactodiglyceride by the method of liposome lysis. However, the significance of this is not clear, since the compound is not present in brain to any extent and 10% of normal controls give positive results (Hirsch and Parks, 1976). Occasional patients also have anti-sulphatide activity in the CSF (Ryberg, 1978). Our own experience in this field has been disappointing as we have been unable to detect increased antibody titres to galacto-cerebroside or ganglioside in MS patients compared with controls. On the other hand, Arnon *et al.* (1980) found anti-glycolipid antibody in about 40% of MS patients using liposome lysis. Activity was reported against GM4 and GM1 and to a lesser degree GM2 and galactocerebroside. It was present in the serum, but not the CSF and appeared to correlate with the severity of the disease. It has also been claimed that galactocerebroside and ganglioside stimulate the formation of 'active' T rosettes *in vitro*, and that this constitutes evidence of cellular hypersensitivity. However, this is a somewhat

indirect way of detecting cellular hypersensitivity and, as little is known of CMI to glycolipids, the report is difficult to evaluate.

12.3.5 Anti-myelin antibody

The simplest way of demonstrating autoantibody in serum is by indirect immunofluorescence and the method is widely used as a screening procedure. However, it is generally held that this technique poses special problems in the CNS and that normal immunoglobulin binds to myelin in tissue sections. This was first reported by Allerand and Yahr (1964) who observed staining of myelin and glia by normal sera. They considered it due to the Fc portion of the molecule and unrelated to specific antibody activity. Subsequently the matter was reinvestigated by Edgington and Delassio (1970) who confirmed that 90% of normal sera reacted with myelin, but claimed that it was due to a naturally occurring IgG antibody which was not present at birth, but acquired in early childhood. Binding was by the Fab and not the Fc fragment and the activity could be removed by specific absorption. Increased titres of this antibody (as well as anti-myelin IgM and A) were found in patients with MS and amyotrophic lateral sclerosis (ALS). These findings were carried a stage further by Lisak *et al.* (1975b) who confirmed that the titre of anti-myelin antibody was raised in MS, ALS and the Guillain-Barré syndrome. These reports do not claim that the immunofluorescent test is of diagnostic value; merely that normal sera have anti-myelin activity and that the mean titre in MS is slightly elevated.

Experience in our own laboratory is completely at variance with the preceding account; we find no evidence that normal immunoglobulin has any special affinity for myelin or that such activity appears in the serum of patients with MS. We have tested a large series of normal and pathological sera on sections of rat and guinea-pig spinal cord, cerebellum and sciatic nerve. Acetone- and alcohol-fixed, as well as unfixed sections were used. No myelin staining could be demonstrated, although 10–15% of normal sera reacted with axons at a dilution of 1:10 giving a ring pattern which might perhaps be mistaken for myelin. Failure to demonstrate myelin staining in alcohol-fixed sections is of particular significance, since treatment with alcohol or a detergent is necessary before the sheath can be stained by specific antisera to MBP.

A careful reading of the earlier reports on the binding of normal immunoglobulin to myelin suggests that the observations are correct but the interpretations misleading, since they failed to take into account the amount of background staining to be expected with undiluted serum. In most routine screening tests the working dilution is 1:8 or more, and a certain amount of non-specific or even specific) binding at higher concentrations is not unusual. For some reason much of the early work on the CNS was done with neat or

relatively concentrated serum and brain came to be regarded as a special case. However, the introduction of potent brain-specific antisera has provided a fresh perspective, and it would seem unlikely that non-specific binding of immunoglobulin in the CNS is greater than in other tissues.

12.3.6 The myelinotoxic factor

In 1963 Bornstein showed that many MS sera were capable of damaging myelin when added to neural cultures in the presence of complement; an observation confirmed and extended by Lumsden (1972). A similar demyelinating factor is present in the serum of animals with EAE and can be demonstrated by its effect on cultures of cerebellum or spinal cord. The earliest change is in the neurological cells, followed by swelling and distortion of the myelin sheaths. These undergo fragmentation and dissolution leaving denuded axons. In some cases lysis of apparently normal myelin may also occur. The myelinolytic factor is present in about 60% of patients with acute MS, but not in inactive cases. It also occurs in amyotrophic lateral sclerosis (ALS) (66%) although the incidence in other neurological diseases is similar to that in controls (8%) (Bornstein, 1965, 1973).

Not all investigators have been able to reproduce this phenomenon. According to Wolfgram *et al.* (1978) there is no difference in myelinotoxicity between MS and normal sera and in some hands the percentage of positives in the general population is 25% (Hughes and Field, 1967). However, these discrepancies are hardly surprising considering the unsatisfactory nature of the test system; myelinating cultures are difficult to maintain and the end-point is highly subjective.

It is generally believed that the toxic factor is an antibody, although the immunoglobulin class and the precise degree of complement is uncertain. According to Dowling *et al.* (1968), demyelinating activity is present in the IgM and IgG serum fractions, although it has proved remarkably difficult to establish this with certainty. The recent work of Grundke-Iqbal and Bornstein (1979) illustrates the elusive nature of this putative antibody. They found it labile and easily inactivated by the usual methods for preparing immunoglobulin. They therefore resorted to the use of staphylococcal protein A, a reagent which binds IgG1, IgG2 and IgG4 and observed that it removed 90–99% of the IgG, leaving most of the demyelinating activity behind in the absorbed serum. Since IgM and IgA were inactive, they concluded that the factor was present in the IgG3 fraction. However, one is left in some doubt as to whether activity, not due to IgM or IgA and only marginally affected by removal of nearly all the IgG, can be due to immunoglobulin at all.

The role of complement is also poorly defined. Although fresh guinea-pig or human serum is usually added, the test serum is not decomplemented in the usual way, as this would reduce its activity. This implies the presence of a heat labile factor, which cannot be restored by the addition of complement

and makes it difficult to determine whether the reaction is complement-dependent or not. The nature of the antigen is unknown apart from the fact that it is present in the CNS. In EAE some of the demyelinating activity may be due to anti-galactocerebroside and MBP is not involved (Seil *et al.*, 1968; Lebar *et al.*, 1976; Dorfman *et al.*, 1978).

Demyelinating activity is present in the CSF although it can only be demonstrated after concentration. Kim and his collaborators (1970) examined pooled samples from patients with MS and found that they demyelinated 80–100% of cerebellar cell cultures compared with 30% when control CSF was used. A similar effect can be produced *in vivo* by injecting unconcentrated CSF into tadpoles in the region of the optic nerve. Demyelinating activity is present in 60% of patients with acute MS and is said to correlate with the severity and duration of the disease (Tabira, Webster and Wray, 1976).

After nearly twenty years the demyelinating factor remains a shadowy and rather insubstantial entity, contributing little to our understanding of the disease. The antigen is unknown and it is not even certain that the effect is due to antibody. Since the factor is present in amyotrophic lateral sclerosis and in a proportion of normal sera, it would seem unlikely that it plays a part in the demyelinating process.

12.3.7 Anti-oligodendrocyte antibody

Since CNS myelin is formed and maintained by oligodendrocytes it is possible that the demyelination in MS might be the result of injury to the supporting cell rather than to the sheath itself. The earliest attempts to demonstrate antibody to glial cells in MS were made by Berg and Kallen (1965). They employed cultures of rat neuroglia as well as cell suspensions and reported gliotoxic activity in 2/3 of MS sera and in animals with EAE. However, it was unrelated to the phase of the disease and present in many other neurological conditions, including head injury.

Methods for isolating oligodendrocytes from mammalian brain are now available, although still relatively crude, and suspensions of these cells have been used for screening MS sera. Abramsky *et al.* (1977) made the striking claim that 19 out of 21 patients had antibody to oligodendroglial surface antigens, detectable by immunofluorescence. They used cell suspensions prepared from bovine brain and obtained similar results in tissue sections. However, this report has received little support from subsequent investigators. According to Traugott *et al.* (1979) immunofluorescent staining of oligodendrocytes is not specific for MS and is due to non-specific binding of immunoglobulin to Fc receptors. MS sera do not bind preferentially to rat oligodendrocytes in culture (Kennedy and Lisak, 1979), nor, in our own experience, do they stain them in sections of brain or sera when examined by indirect immunofluorescence.

transmission has much to commend it. However, the methods used are subject to great biological variation and the results are difficult to evaluate. There is still no hard evidence that these factors— if they exist at all—play a significant role in the pathogenesis of the disease.

12.4 THE ROLE OF IMMUNITY IN PATHOGENESIS

12.4.1 · Is MS an 'immune disorder'?

In most discussions on MS it is assumed that immunological processes are involved in its pathogenesis. In the sense that all inflammatory and infectious diseases have an immune component, this is undoubtedly true. However, the argument is frequently carried a stage further with the suggestion that the disease is some form of immune disorder, but the evidence on this point is much less convincing. The precise nature of the proposed disturbance is often a little vague, but there appear to be three main suggestions. The first is that the immune status of the MS patient is abnormal, and that fluctuations in immune reactivity are responsible for the characteristic disease pattern with its exacerbations and remissions. The second is that the immune response in the CNS represents an aberration of local immunity and that there is an immune defect underlying the persistent infection. Finally, there is the view that the demyelination is not due to the direct effect of a virus or toxin, but to autoimmunity or hypersensitivity. It may be that some—or all—of these propositions are true, but in each case the evidence is inconclusive. It is at least arguable that, on present evidence, MS is no more an immune disorder than neurosyphilis, a disease in which immunoglobulin is synthesized intrathecally, and which is diagnosed by the presence of autoantibody in the serum and CSF.

The immunological profile of patients with MS is set out in Table 12.1. It includes a minimal reduction in cell-mediated immunity (CMT) and fluctuations in the Tγ, T_S and T_A subsets of the peripheral blood. Although of great interest, these findings do not satisfactorily explain the two main problems of systemic immunity in MS, namely susceptibility to the disease and the remitting pattern of the pathological process. The depression of CMI is a marginal and inconstant feature of the illness and is also present in neurological controls. It appears to be the result of long-standing disease in the CNS and cannot be regarded as evidence of a primary immunological disorder; nor is it likely to play a significant role in the pathogeneis. The same goes for the depression of 'active' T-lymphocytes, which is present in many infections and may be regarded as a normal concomitant of viral disease.

Variations in the suppressor cells may be of greater significance, and it is tempting to try to explain disease activity in terms of an immune process released from the constraints of the normal control mechanism. One suggestion is that the reduction in the number of Ts cells in the peripheral

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CHAPTER THIRTEEN

Multiple Sclerosis and Chronic Relapsing EAE: Comparative Ultrastructural Neuropathology

Cedric S. Raine

13.1 Introduction
13.2 Multiple sclerosis
13.3 Chronic relapsing experimental allergic encephalomyelitis
13.4 Conclusions

13.1 INTRODUCTION

13.1.1 An immunological rationale for the genesis of the MS plaque

Regardless of the nature of the putative aetiological factor in multiple sclerosis (MS), in the face of an increasing literature on immunological anomalies, it is more than likely that immune-mediated events play a significant role in the expression of the ensuing disease within the central nervous system (CNS) in this disorder. That these immunological vagaries and their fluctuations might herald pathogenetic events provide the scaffold for the following paragraphs which will attempt to support the hypothesis by ultrastructural data. While the variation in clinical picture innate to MS (Hallpike, Chapter 11), renders correlation of findings from the various immunological parameters hazardous, the morphology of demyelinating lesions in MS is best appreciated when presented against the backcloth of the growing evidence for an immunogenic process. In this context and supporting the possibility of a generalized immunological abnormality in MS, a large number of works exist. The following represent but a few on this subject. Kabat *et al.* (1950) first detected an elevated IgG level in the cerebrospinal fluid (CSF) of a significant number of MS subjects, a finding suggestive of local antibody synthesis. Link (1972) and Johnson *et al.* (1977) observed oligoclonal IgG in the CSF of MS subjects, indicative perhaps of some specificity in the CSF antibody responses. Later, Williams *et al.* (1978) showed IgM in the CSF of a significant percentage of MS subjects. IgM usually represents a primary antibody response to infection or persistence of antigen. Within the circulation, Oger *et al.* (1975), Santoli *et al.* (1978), Kam-Hansen,

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CHAPTER FIFTEEN

Immunoregulatory Mechanisms in Experimental Autoimmune Encephalomyelitis and Multiple Sclerosis

C. C. A. Bernard, P. R. Carnegie and I. R. Mackay

15.1 Introduction
15.2 The model disease, experimental autoimmune encephalomyelitis
15.3 Immune dysregulation in multiple sclerosis
15.4 Conclusions

15.1 INTRODUCTION

Experimental autoimmune encephalomyelitis (EAE) is widely regarded as a laboratory model for multiple sclerosis (MS) (Cuzner and Davison, 1979; Eylar, 1979; Kies, 1978; Paterson, 1977). Acute EAE as seen in the guinea pig and rat is closer to human post-vaccinal encephalomyelitis than to MS, whereas chronic EAE as seen in the guinea pig shows neuropathological damage resembling that seen in MS (see Chapter 13 by Raine). The recently increasing evidence of immune dysregulation in MS points to the need for detailed experimental study of immunoregulatory processes in both acute and chronic EAE, but progress will be slow because the immune system is highly complex (Fig. 15.1). There are factors identified which regulate the development of the various classes of lymphocytes in the antigen-independent phase, and others operative in the antigen-dependent phase. However, there are now several sites on various arms of the immune response where abnormalities in regulation have been identified, and several sites at which drugs can influence this response (Fudenberg *et al.*, 1980), and it is opportune to apply current knowledge in this area to the question of the pathogenesis of

15.2.1 Historical background

The concept that tissue of the central nervous system (CNS) has unique antigenic constituents and can provoke an autoimmune response can be

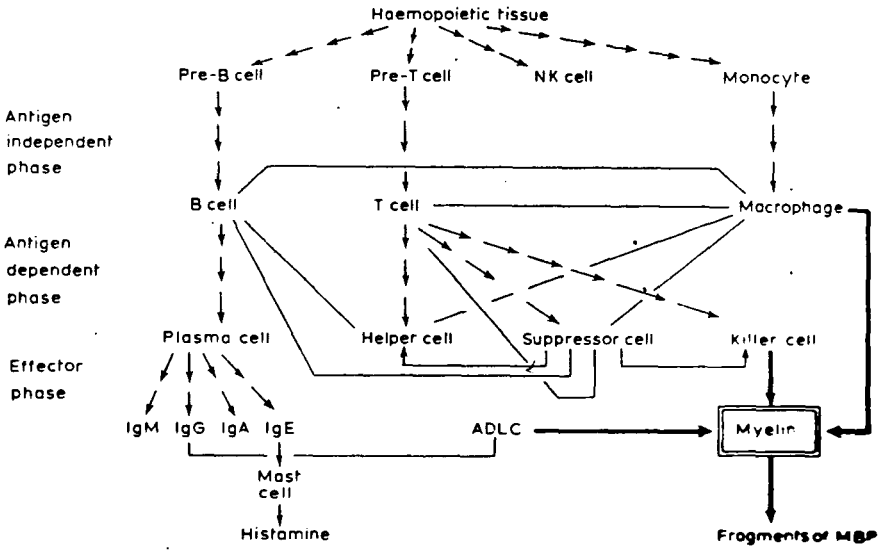

Fig. 15.1 Complexity of immune system in relation to autoimmune damage to myelin. Myelin can be damaged by lytic enzymes released or activated by lymphocytes or macrophages. Myelin can also be damaged by an immune response to a non-myelin component as a result of release of lytic enzymes (bystander damage).

traced back to the time of Louis Pasteur, when neuroparalytic accidents were observed after injection of rabies vaccine prepared from the spinal cord of rabbits. Historically, studies on CNS have been highly prominent in providing concepts on self-tolerance; self-reactivity and mechanisms of autoimmunity. Indeed, one of the first organ-specific tissue antigens to be recognized as producing specific autoimmune responses was central nervous tissue. The observation that injection of brain and spinal cord material could induce neurological disease in animals, experimental autoimmune encephalomyelitis (EAE), provided one of the first laboratory autoimmune model systems and established the reality, contrary to the concept of 'horror autotoxicus' (Ehrlich, 1900), that immune responses to self antigens could cause an immunological and pathological disease. An important step in understanding the nature of EAE has been the definition of CNS antigens responsible for disease induction. Rivers and Schwentker (1935) reported that acute encephalomyelitis induced in monkeys by nervous tissue immunization closely resembled post-rabies vaccinal encephalomyelitis. Impressed by the degree of demyelination present in such immunized animals, Rivers, Sprunt and Berry (1933) and Rivers and Schwentker (1935) suggested that the immunological responses to nervous tissue antigen could be related to the profound degree of myelin injury so characteristic of MS. The introduction of Freund's complete adjuvant (FCA) in the early 1940s greatly facilitated the induction of EAE in many animal species (Freund, Stern and Pisani, 1947).

Indeed, a single injection of nervous tissue emulsified in FCA regularly induced within 2 to 3 weeks an accelerated and extremely severe form of disseminated encephalomyelitis in guinea pigs, monkeys and rabbits. The autoimmune character of this disease in experimental animals was established by Kabat, Glusman and Knaub (1948) showing that monkeys developed EAE after sensitization with autologous cerebral tissue. Since then, many reports describing a variety of encephalitogenic preparations which induced disease in different species have appeared in the literature, reviewed by Kies (1973) and Paterson (1977).

Important for understanding the immunological reactions was the eludication of the structure of the encephalitogenic agent and its location in the CNS. After considerable confusion as to the nature of the encephalitogen and debates between groups led by Kies, Einstein and Lumsden on the size of the molecule, it was eventually established that the encephalitogen was myelin basic protein (MBP) (Kies, 1965). Subsequently, a key finding was that MBP could be readily digested during isolation without destruction of the encephalitogenic region (Carnegie, Bencina and Lamoureux, 1967; Nakao, Davis and Roboz-Einstein, 1966), and relatively low activity of the other CNS fractions was shown to be due to contamination with MBP or peptides derived from it (Kies, 1973).

15.2.2 Myelin basic protein

MBP has been isolated from man, rabbit, guinea pig, chicken, monkey, horse, sheep, dog, turtle and frog (Deibler, Martenson and Kies, 1972; Dunkley and Carnegie, 1974). Myelin from each of the above species contains a single basic

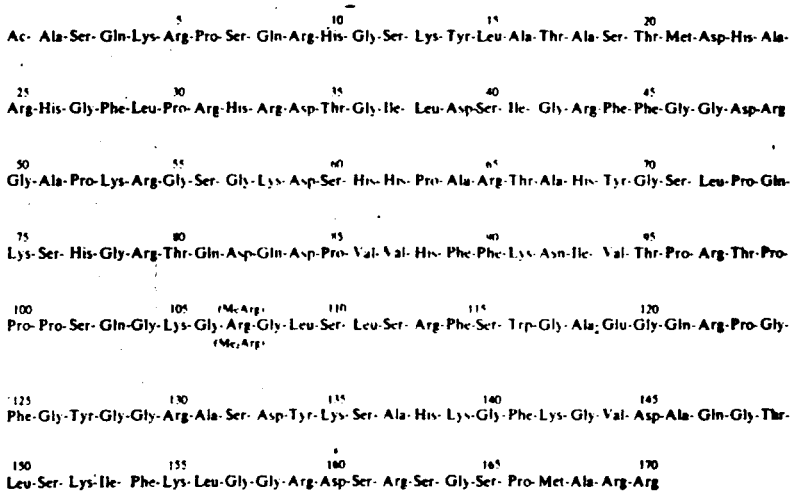

Fig. 15.2 Amino acid sequence of human myelin basic protein (Carnegie, 1971a).

protein with similar electrophoretic and chromatographic properties (Carnegie and Dunkley, 1975). However, not all species contain a single MBP in that the rat, mouse, and other rodents in the *Myomorpha* and *Sciuromorpha*, e.g. hamster and squirrel, have an additional smaller MBP (Bernard and Carnegie, 1975; Carnegie and Dunkley, 1975; Martenson, Deibler and Kies, 1971). This smaller MBP has an unusually long deletion of some forty amino acids in a continuous sequence (Dunkley and Carnegie, 1974). The amino acid sequence of MBP from man (Fig. 15.2) (Carnegie, 1971a), ox (Eylar *et al.*, 1971) and rat (Dunkley and Carnegie, 1974) has been determined. A characteristic feature of this protein is its high content of the basic amino acids lysine, histidine and arginine, which represent over 25% of the residues. Current knowledge on the structure and localization of MPB in myelin has been reviewed by Carnegie and Moore (1980).

15.2.3 Other candidate encephalitogenic antigens

There has been continuing interest in components of brain other than MBP as possible determinants of EAE, although the studies have not reached high levels of conviction. Worthy of mention are reports of a cerebroside which induces antibody with anti-glial and demyelinating activity *in vitro* (Dubois-Dalcq, Niedieck and Buyse, 1970; Fry *et al.*, 1974), a non-cerebroside which induces antibody with demyelinating activity (Lebar *et al.*, 1976), and a hydrophilic lipoprotein of myelin (lipophilin) which is encephalitogenic and induces cell-mediated immunity in immunized guinea pigs (Hashim, Wood and Moscarello, 1980).

15.2.4 Microheterogeneity in MBP

Myelin basic protein, as isolated in the laboratory, is in reality a collection of protein molecules which differ from each other as a result of enzymic modifications after protein synthesis (Table 15.1). The function of these modifications is not understood but it is possible that some of them are involved in diseases of myelin. While it is not possible to separate completely all of these modified forms, it is possible partially to resolve MBP by ion-exchange chromatography at pH 10.6 (Chou *et al.*, 1977). The more acidic forms can be separated from MBP but there is no known way of separating the methylated forms of MBP from each other. Thus, within the apparently sharp peaks from the exchange column, a heterogeneous mixture remains. Two of the enzymic modifications, methylation and phosphorylation, will be examined in more detail because of their possible relevance to demyelinating diseases.

15.2.4(a) Methylation

Arginine-107 in MBP can occur in its unmodified form or as mono-

Table 15.1 Enzymic modification of MBP

Modification	Site	Comments
Acetylation	*N*-terminal Alanine-1	Protection from peptidases
Methylation	Arginine-107	See text
Phosphorylation	Several sites	See text
Glycosylation	Threonine-98	Demonstrated *in vitro* not located as yet *in vivo*
Deamidation	Glutamine-103	May be involved in degradation
Removal of arginine	*C*-terminal Arginine-170	May only occur during isolation

(For refs. see Carnegie and Moore, 1980)

methylarginine or dimethylarginine (Baldwin and Carnegie, 1971). The extent of methylation of MBP varies from species to species, but is constant within adults from one species. For example, in the human approximately 94% of arginine-107 is methylated and in the rat 27% (Martenson, 1980). Methylation of this residue prevents the action of trypsin-like enzymes at this site. Methylation may be involved in getting this region of the molecule into a more hydrophobic environment (Littlemore and Ledeen, 1977). Recently, we (Small, Carnegie and Anderson, 1981a) have studied the methylation of MBP *in vivo* and presented evidence that inhibition of methylation of MBP might be involved in subacute combined degeneration of the spinal cord, a disease due to prolonged vitamin B_{12} deficiency. The large vacuoles in myelin found in this disease suggest that the lamellae are not properly knitted together. If animals are treated with cycloleucine identical lesions are produced and the methylation of MBP is inhibited (Small *et al.*, 1981a). From this and other evidence (Small, Carnegie and Stuart, 1981b), it is now becoming clear that vitamin B_{12} is required for the reformation of methionine in the CNS and a deficiency of vitamin B_{12} will lead to a failure in the methylation of MBP (Crang and Jacobsen, 1980; Small *et al.*, 1981b).

15.2.4(b) Phosphorylation

A curious feature of MBP is that a small proportion (10–20%) of the molecules are phosphorylated (Chou *et al.*, 1977; Martenson, Kramer and Deibler, 1976). The localization and function of phospho-MBP is not known, but it could have an important role in myelin function. *In vivo*, at least three enzymes are involved in phosphorylation and dephosphorylation of MBP, a cyclic AMP-dependent protein kinase located in the cytosol, a myelin associated kinase (Carnegie *et al.*, 1974; Carnegie, unpublished work), and myelin phosphoprotein phosphatase (Miyamoto and Kakiuchi, 1975). The myelin kinase is of particular interest as it is activated by small changes in the

IgG in MS CSF are synthesized within the brain CSF compartment, which is consistent with observations that active plasma cells are frequently found in the vicinity of MS plaques (see Esiri, 1980 and Adams, Chapter 8, for further information).

The nature of the antigenic stimulus for increased production of IgG in CSF and CNS is unknown: normal brain components may be implicated or one or another virus, or there may be random activation of 'irrevelant' B cells in the neuraxis. Part of the increased IgG is directed against measles antigens (Mehta, Thormar and Wiśniewski, 1980) and part against MBP (Bernard *et al.*, 1981a; Panitch, Hooper and Johnson, 1980), but antibody to these particular antigens does not by any means account for all of the increased IgG in CSF. The existence of raised levels of IgG in CSF in MS takes on added relevance from recent studies on immunoglobulin extracted from the brains of patients with MS at autopsy (Bernard *et al.*, 1981a, b). Such brains contained greatly increased amounts of IgG, with limited and discrete patterns of Ig banding as is seen in CSF (Mattson, Roos and Arnason, 1980), of which a substantial proportion was antibody to MBP and, whilst some antibody to measles and other viruses was demonstrable, no cross-reactivity between antibody to MBP and viruses could be detected (Bernard *et al.*, 1981b). It is an attractive possibility that the monoclonal bands in CSF from different patients with MS contain common idiotypes but at present there is no clear evidence for this (Arnon *et al.*, 1979; Baird *et al.*, 1980; Nagelkerken *et al.*, 1980). In three patients with subacute sclerosing panencephalitis (SSPE) there were similar spectrotypes in CSF on isoelectric focusing (Mattson *et al.*, 1980), and hence search for common idiotypes could be more successful in SSPE than in MS.

15.3.3 Immune-mediated myelinosis

MS is the prototype of the 'demyelinating' diseases, yet the mechanisms of myelin breakdown are still imperfectly understood. In emphasizing that MBP, the target antigen of EAE, has a key role in maintaining the structure of myelin, we draw attention to suggested models for the orientation of lipids and proteins in myelin, one example of which is shown in Fig. 15.3.

First of all, increased proteolytic activity at the edge of active MS plaques could account for breakdown of myelin (Einstein *et al.*, 1972), since MBP is susceptible to digestion by various proteinases, in contrast to the relative resistance of the other main myelin protein, proteolipid protein (see also Hallpike, Chapter 11). Cuzner and Davison (1979) found in CSF from patients with MS that there were increased levels of proteolytic enzymes, presumably originating from lymphocytes and macrophages around the plaques. Norton *et al.* (1978) demonstrated that macrophages, in addition to releasing degradative enzymes, could activate the serum enzyme plasmin which rapidly digests MBP in myelin.

Studies on proteolysis of myelin explain how myelin might be damaged by

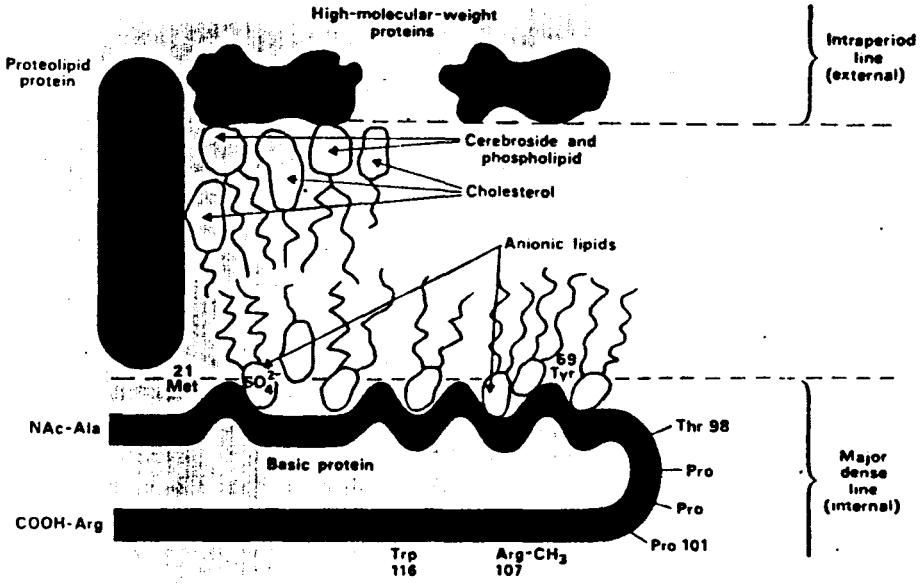

Fig. 15.3 Hypothetical model of myelin structure which shows a possible relationship between the lipids and proteins in the myelin lamellae. (Reproduced from Davison and Cuzner, 1977.)

an immune response to a non-myelin component. For example, if a virus or a viral antigen expressed at the surface of a glial cell were to stimulate an immune response, the resultant release of proteolytic enzymes could lead to a breakdown of adjacent myelin because of its high susceptibility to enzymic digestion, the 'bystander' effect described by Wiśniewski (1977) and discussed in Chapter 8. The demyelination observed in Theiler's disease, an encephalomyelitis of mice, may provide an example of this type of 'bystander' demyelination, for which an intact immune system is required. It is of interest that the extent of demyelination in Theiler's disease appeared to correlate better with the presence of macrophages than with the amount of virus (Lipton and Dal Canto, 1979).

The question of proteolytic damage to myelin leads on to the several reports on the presence of MBP or fragments of MBP in the CSF of patients with MS (Cohen *et al.*, 1976; Whitaker, 1977) and other diseases (*vide infra*), suggesting that proteolysis of myelin does occur early in exacerbations. Similarly, vulnerability of myelin to proteolytic damage explains findings on immunoassay for MBP molecules in plasma immediately after cerebrovascular accidents (Palfreyman *et al.*, 1979) and head injuries (Thomas, Rabow and Teasdale, 1979), and in the CSF after brain surgery (Alling, Karlson and Uällfors, 1980); whether these molecules represent intact MBP or large antigenic fragments remains to be determined. In addition, the question

is quite open as to whether release of MBP is more likely to be a provocative or suppressive stimulus of an immune response to MBP.

Electron microscopy portrays myelin as quite a stable and highly ordered structure, but *in vivo* the opposite is the case. Myelin is a particularly vulnerable membrane, more so than most other membranes, as evidenced by the large vacuoles which appear in response to quite small amounts of chemical toxins such as hexachlorophenes (Cammer, Rose and Norton, 1975) and similar damage occurs readily when methyl group metabolism is disrupted by vitamin B_{12} deficiency, treatment with cycloleucine or nitrous oxide intoxication (Small *et al.*, 1981a, b).

We now suggest the drawing together of two concepts, namely 'bystander' damage to myelin and vulnerability of myelin to proteolytic damage *in vivo*, in relation to the formation of MS plaques, and new approaches to treatment of MS. Thus in the genesis of plaques, the effects of antibody to neural components, particularly MBP, and possibly virally-coded antigens expressed on neural cells, could generate conditions predisposing to 'bystander' damage to various components, but particularly to the highly vulnerable myelin sheath. In regard to new approaches to treatment, we can refer to nutritional regimes aimed at maintaining myelin in a stable state, e.g. unsaturated fatty acids which reportedly are beneficial (Field, 1980), and inhibition of proteolytic enzymes, a procedure recently shown to interfere with development of EAE (Brosnan *et al.*, 1980), and worthy of trial to minimize plaque formation in MS.

15.3.4 Changes in lymphocyte populations

The availability of markers for defining human lymphocyte populations has resulted in increased attention being paid to the role of subpopulations of T cells in various immunopathic diseases, including MS. Fluctuations in number of T cells have been observed in peripheral blood and CSF of patients with MS (Merrill *et al.*, 1980; Traugott, 1978) and, in particular, there is a diminution in numbers in blood of 'active' T cells defined by E rosetting (Kam-Hansen, 1979; Kately and Bazzell, 1979). Moreover, natural killer lymphocytes in MS display a significantly lower killer activity as compared with those of healthy subjects, and have an impaired response to interferon and interferon inducers (Benczur *et al.*, 1980). Of special interest, patients with 'active' MS show a striking alteration in numbers of T suppressor (Ts) cells, indicative of faulty immune regulation. As circumstantial evidence for this, we may cite the well-known evidence that viral antibody levels are increased in blood and CSF in MS. Direct evidence for abnormalities in function and/or numbers of Ts cells in MS is now quite substantial, and includes (a) deficient *in vitro* activation of suppressor lymphocytes with Con A (Antel, Weinrich and Arnason, 1978; Wicher, Olszewski and Milgrom, 1979); (b) altered numbers of cells with Fc receptors for IgG (Ty cells), increased in most cases according to Santoli *et al.*

CHAPTER SIXTEEN

Comprehensive Management of Multiple Sclerosis

Wallace W. Tourtellotte and Robert W. Baumhefner
with the assistance of
Janet H. Potvin, Alfred R. Potvin and S. Poser

16.1 Introduction
16.2 Informing the patient of the diagnosis and prognosis
16.3 Responding to common inquiries
16.4 Planning an exercise programme in addition to daily tasks of living
16.5 Treating neurological symptoms and signs
16.6 Preventing and treating secondary complications
16.7 Putative treatments
16.8 On keeping current

16.1 INTRODUCTION

Approximately one and a half million people in the world now suffer with Multiple Sclerosis (MS),* and in each patient the manifestations of the disease vary, requiring different management approaches. The specific cause of MS is still unknown, and there is no generally accepted therapy of proven value. While any hope for eradication of the disease, either by prevention or by specific therapy, requires an understanding of its aetiology and pathogenesis (Bauer, 1977, 1978), effective management requires knowledge of symptoms, signs, and problems and appropriate methods of dealing with them. In this chapter, a compilation of existing information about managing and treating MS is presented.

The combination of sensory, motor, and coordinative impairment is unique to each MS patient and varies in each from time to time (Slater and Yearwood, 1980). Unpredictable exacerbations and remissions occur in a majority of patients. The clinical course of the disease varies remarkably from patient to patient as does the severity of the impairment in each relapse that a patient suffers. Fatigue (accentuated by heat) and depression can considerably worsen functional ability, falsely indicating disease progression

* This calculation is based on 4×10^9 people in the world with 1×10^9 at high risk of $80/10^5$ and 3×10^9 at low risk of $10/10^5$ Kurtzke, 1980).

Europe and North America (reviewed by Kurtzke, 1980 and Kurtzke, Chapter 3) indicate that there is a geographical distribution for the frequency of MS which varies with latitude. A high prevalence of MS (30–80 per 100 000 population) is found above 40° N latitude and a lower prevalence (less than 5 per 100 000) is found below 12° N. Some sort of environmental factor seems likely, because migration from a high- to low-risk area and vice versa can cause the prevalence for MS in migrant populations to be more like that of the area migrated to. However, the age at which one migrates is critical; it must be prior to the age of 15. Studies tend to demonstrate that after the age of 15, the migrant carries with him/her some, but not all, of the risk of the region migrated from (Visscher *et al.*, 1977; Kurtzke, Chapter 3).

16.3.5 Heat sensitivity

For many, but not all patients, exposure to heat has a temporary adverse effect on MS symptoms and signs. It is common to feel worse during hot and humid weather and to experience increased disability as a result of any increase in body temperature, such as that caused by a fever or a hot bath. These changes in function due to raising the body temperature 0.5°C have been confirmed in the laboratory (Davis, 1966; Davis, Michael and Neer, 1973). In addition, visual evoked potentials can be made worse (Saul and Selhorst, 1981).

We believe that exposure to heat for a short time does not cause a worsening of the disease itself, but to those patients affected by heat we advocate the utilization of air conditioners as much as possible, air conditioning at least one room (preferably the bedroom) at home and a room at work as well as the car. (With proper documentation from the physician, air conditioning costs can be a tax-deductible expense in the USA.) We also have the patients avoid prolonged exposure to the sun, confine their most vigorous activities to the cooler part of the day, avoid overdressing, and take lukewarm or cool showers.

16.3.6 Dental care with or without regional anaesthesia

We have not been impressed by the adverse effects on the course of MS from any dental procedures, e.g. fillings of all types and multiple extractions. Evidence in the literature is insufficient to warrant even a policy of moderation (Firnhaber and Orth, 1977). Local anaesthesia has not been found to increase the number of relapses above that expected for the natural history of the disease (Bamford, Sibley and Laguna, 1978a).

16.3.7 Immunization

In the past (McAlpine *et al.*, 1972) physicians tended to avoid innoculation of MS patients with any antigens. However, in recent years, two studies (Myers *et*

al., 1976; Bamford *et al.*, 1978b) have indicated that no significant difference in the relapse rate and increased disability exists between a group of MS patients vaccinated with swine influenza-Victoria A vaccine and a control group of MS patients. Accordingly, there exists some support for preventive medicine utilizing a specific influenza immunization.

Bauer (1977) stated that for practical purposes, the urgency of vaccination should be weighed against a possible risk in individual cases. Except for smallpox and possibly poliomyelitis, in his experience, there exists no danger for yellow fever, cholera, tetanus, measles, influenza, mumps and rubella if the patient is not having a bout of pronounced clinical activity. He also advised that steroid administration should be avoided two weeks before and after any vaccination.

Nevertheless, we do not recommend routine immunizations of our MS patients. In a recent report (Walsh, Tourtellotte and Potvin, 1982) we have postulated that one of the pathological aspects of a patient with clinically definite MS is the presence of lymphatic tissue in the CNS, a tissue which is not normally present. Because of that, when the body is confronted with an antigen, such as that which occurs with immunization, all lymphatic tissue of the body is activated which should include the pathological lymphatic tissue in the CNS. Accordingly, conditions in the CNS could be precipitated that would lead to a worsening. In addition, some antigens in vaccines produce a high fever for a few days which could in itself cause a serious worsening manifested by dysphagia and possible pulmonary aspiration, especially if the patient has evidence of brain stem lesions.

16.3.8 Infections with or without fever

Severe infections are obviously deleterious to an already ill person. In MS, if a fever develops it is common to recapitulate old symptoms and signs and to worsen existing ones. On the average, a worsening of the neurological condition will last only for a day or two after the body temperature has returned to normal. We refer to this as a fluctuation in the neurological condition owing to the fever; it is not a relapse. However, infections may precede the onset of a significant number of relapses (Miller, 1961; Scheinberg, 1979).

We consider hyperpyrexia in a MS patient a neurological emergency. It is necessary to prevent a recapitulation or worsening of existing disabilities induced by hyperthermia such as dysphagia and subsequent pulmonary aspiration. Accordingly, treatment immediately with antipyretics (aspirin or acetaminophen) is instituted. If this fails to reduce the temperature, a cooling blanket, alcohol soaks, or cool bath is indicated. Immediate treatment of infections with broad spectrum antibiotics after cultures have been taken is our practice. Changes in antibiotic prescriptions are made as the sensitivity of the micro-organisms isolated are identified.

16.3.9 Injuries severe enough to require restriction of activity

Whether physical trauma (sprains, fractures, concussions, burns), irrespective of site or magnitude, can contribute to precipitation of MS attacks remains controversial (McAlpine *et al.*, 1972). Even though MS patients should be expected to be more subject to accidents than normal persons because of their motor or visual disabilities, experience does not bear this out. It is possible that MS patients, because of their neurological disabilities, are subconsciously more cautious, and hence avoid hazardous situations.

When an accident occurs and a period of confinement to bed is prescribed, increased difficulty in getting about often follows. We recommend that every effort be made to keep rest as a treatment to a minimum; rest in a chair might be a compromise. If bed rest or chair rest is necessary, active and passive exercises should be carried out twice daily under the direction of a physiatrist or aide. (See also Section 16.3.16—Fatigue caused by daily tasks of living and rest periods.) If prolonged bed rest is necessary, the risk of pulmonary emboli can be minimized by daily administration of low-dose heparin.

16.3.10 Pregnancy, contraception, abortion

The effect of pregnancy on the course of MS is unpredictable. It has been stated that pregnancy has no effect on the course of MS if the patient has had no exacerbation for two years (Scheinberg, 1979). On the other hand, symptoms of MS may begin with or be accentuated during pregnancy or in the weeks post partum (McAlpine *et al.*, 1972). A precise study (Schapira *et al.*, 1966) of females of child-bearing age has shown that the risk of exacerbation is small (0.25 exacerbations per 'pregnancy year', i.e. 9 months of pregnancy and 3 months post partum), but modestly greater than in a 'non-pregnancy year' (0.16 per year). The number of relapses in the 3 months following delivery was equal to the number in the 9 months of pregnancy. Hence, on average MS patients have less worsening while carrying the baby, but catch up in the post-partum period. This is in agreement with Poser (1982). In addition it was found that MS itself did not influence the normal course of pregnancy and childbirth. Schapira *et al.* (1966) found that women who had the onset of MS after pregnancy were more disabled than female patients without pregnancies. Therefore, they concluded pregnancy constitutes a calculated risk for the patient with MS, and must be taken into account in counselling. On the other hand, Millar *et al.* (1959) and Poser (1982) reported no significant differences in prognosis between pregnant and non-pregnant MS patients.

Based on the above statistics and our experience we believe that the most important issues in deciding to have children are practical matters. At the time of conception the patient should have the motivation, intelligence, and stamina to care for an infant and then an active child. Does the family have a back-up plan in case the mother becomes unable to care for the child full

time? Are relatives or friends nearby from whom regular assistance can be obtained, or is there enough money to hire someone to help?

Unquestionably, caring for an infant and an active child when the mother has a severe physical disability leads to frustration and causes deleterious effects both in patient and child, even under the best of supportive programmes.

Oral contraception, is well tolerated by MS patients (Poser *et al.*, 1979). In our experience tubal ligation and abortion have presented no problem.

16.3.11 Obstetrical anaesthesia

The method of delivery should depend on the indications. Any type of anaesthesia may be used, although based on a recent review (Bamford *et al.*, 1978a) there may be less of a problem with general anaesthesia than with

In our experience and that of others (Bamford *et al.*, 1978a; Miller, 1961; Ridley and Schapira, 1961) indicated surgical operations should be carried out. The choice of anaesthesia should be made on the basis of the surgical procedure to be performed and the experience of the anaesthetist. From their clinical experience and a review of the literature, Bamford *et al.* 1978a) concluded that spinal anaesthesia is less preferable than general anaesthesia. They do not encourage spinal anaesthesia in MS patients if a reasonably safe and convenient alternative is available. General anaesthesia caused no increase in the relapse rate of MS above that expected in their study, and their review of the literature for the most part supported this observation.

16.3.13 Other diseases including brain tumours

A recent post-mortem study (Allen, Millar and Hutchinson, 1978 of 120 necropsy proven cases of MS showed no significant increases in the incidence of myocardial infarction, cerebral infarction, cerebral haemorrhage or malignancy compared with matched patients having other neurological disease or general controls. Seventy-four per cent of cases died from complications of pulmonary and renal infections.

Several reports of astrocytomas or glioblastomas in MS (Currie and Urich, 1974; Spaar and Wilkström, 1978; Kalimo *et al.*, 1979) have raised the possibility that these tumours may arise from reactive astrocytes in the plaque tissue, but such occurrences are extremely rare. We have not encountered such a case, even though we screen all MS patients with an enhanced CT brain scan.

16.3.14 Emotional stress and disorders

While patients with well-developed MS symptoms may show emotional instability that is probably the result of organic changes in the brain

(Langworthy, 1948), patients for the most part exhibit normal emotional reactions associated with their illness and maintain intact thinking processes.

When first confronted with the diagnosis, the MS patient usually goes through several stages of adaptation (Yearwood, 1980a). The first stage is often denial; an attempt may be made to conceal symptoms and to seek out a health-care professional who will help refute the diagnosis. The second stage of adaptation is characterized by resistance, a constant fight against the disease, a fierce independence, and an unwillingness to accept help. Trips may be made to several centres looking for 'the cure' to the disease. The next stage may be thought of as affirmation. The patient begins to learn to face the diagnosis and accept help. Finally, the stage of integration is reached when the patient accepts the fact that she/he is not the same person, but can continue to live. At this stage, the disease is fully accepted and is not constantly in the mainstream of thought. It is important to help guide MS patients into full acceptance of their disease; however, there is often ambivalence by physicians and nurses toward MS patients and vice versa that can interfere. Indications of resentment towards MS patients because they fail to respond to treatment, although understandable, must be resisted. The lines of communication must remain open and an active effort made to alleviate the patients' minor complaints, many of which may be of great importance to them (*Multiple Sclerosis: Guidelines for Diagnosis and Management* 1980). Patients should be told to report by telephone any problems they are having and to report at least annually for a neurological evaluation. Group therapy sessions, in which patients with similar disabilities discuss their mutual problems and provide support for each other, are effective in enabling patients and their families to adapt to the disease. Programmes of this nature are available at almost every chapter of the National Multiple Sclerosis Society as one facet of the broad scope of patient service programmes that it sponsors (Schneitzer, 1978).

Social and psychic tensions are facts of life and it is not possible to protect MS patients from them; however, certain crisis situations can be anticipated. MS often strikes the patient in the prime of life, in the midst of raising a family and establishing career goals, which causes enormous adjustment problems for the patient, the family, and others who come in contact with him/her (Putton, 1977). Often marriages cannot bear up under the strain and divorce ensues. The patient's self-image is compromised and he/she is sometimes acutely concerned with future loss of somatic or intellectual function. The patient may still be self-sufficient but may lose employment because of lack of understanding by the patient's employer. The neurologist can discuss MS with the employer and suggest how job requirements may be modified so that the patient can continue to work. Anticipatory and aggressive counselling and education of the patient, his/her family, and his/her employer and social contacts can head off these stressful situations.

A recent study by Baretz and Stephenson (1981) revealed that the majority of MS patients evidence concealed depression; overt depression is the second

most predominant reaction. Although MS patients have been characterized as euphoric (McAlpine *et al.*, 1972) their study did not show the patients as having a high rate of elevated moods. With progression of the disease, overt depression tended to increase, while denial seemed to decrease. Baretz and Stephenson (1981) recommended helping the MS patient cope with depression as a major therapeutic goal.

Psychiatric counselling should be available not only to the patient but also to the family which may experience a difficult time coping with the changes in lifestyle and member relationships inevitable with this disease. Only occasionally are drugs indicated such as the CNS stimulants dextro-amphetamines (Dexedrine™) or meythylphenidate (Ritalin™), or the antidepressive drugs imipramine (Tofranil™) or amitriptyline (Ellavil™). Drugs are not a substitute for verbalizing interpersonal relationships.

Several reports (Langworthy, 1948; McAlpine, 1957; McAlpine *et al.*, 1972) indicate that psychological stress can lead to the onset of the first symptoms of MS or contribute to exacerbations in patients with established disease. We believe this is controversial, but emotional stress can sometimes be associated with a temporary worsening in much the same way that a worsening can be associated with fever or excess fatigue.

Recently, Miles *et al.* (1981) studied the sympathetic nervous system as a regulator of the immune response in mice. Their findings indicated that modulation of the cellular immune response can be produced by sympathectomy or axotomy with 6-hydroxydopamine. Further studies on the new discipline of psycho-immunology may relate to relapses and fluctuations in MS with the immune system via the autonomic nervous system.

16.3.15 Sexual problems

Sexual dysfunction is quite common in MS. In one report (Lilius, Valtonen and Wilkström, 1976) it was found that sexual life was changed in 91% of the men and 72% of the women with about half the patients having either an unsatisfactory or discontinued sexual life. Impotence may be an early symptom of MS. In the course of the disease, most male MS patients have some degree of impotence, which can become complete. It is manifested by inability to achieve or sustain an erection or to ejaculate, or both. Sometimes ejaculation may become retrograde into the bladder. Libido is only slightly decreased in most cases. Women complain of inability to attain orgasm, loss of libido and interference with the sexual act because of spasticity. Muscle relaxant drugs that relieve spasticity may occasionally be beneficial. Sexual counselling should be available to all patients. A significant percentage of sexual problems in MS is still related to psychological causes; accordingly, these patients can benefit from counselling. In addition, spouses of patients should be informed of the nature of the problem and should receive instruction on compensatory methods in sexual intercourse (Schneitzer, 1978).

The evaluation of impotence in the male should include a nocturnal penile tumescence recording and, if indicated, a penile prosthesis should be considered. The best prosthesis employs inflatable silastic rods implanted in the corpora cavernosa, and gives an erection at the desired time (Furlow, 1980).

16.3.16 Fatigue caused by daily tasks of living and rest periods

Fatigue frequently accompanied by weakness to the point of exhaustion is present at some time in most MS patients (McAlpine *et al.*, 1972). It is often inexplicable on the basis of the neurological findings and may occur even after a good night's sleep in patients who have normal or only minimally abnormal neurological examinations. Patients must be encouraged not to give in to fatigue. Few patients will be able to carry on as they normally do when they feel this way, but they should try. They will often prove to themselves that the more they attempt the easier it becomes, improving their morale and gaining a feeling of having some control over their condition.

On the other hand, over-exhaustion may lead to a temporary increase in symptoms and should therefore be guarded against.

We are not impressed with the value of bedrest as part of the treatment for acute exacerbations, nor is Russell (1976). Patients with MS should be active and busy during the day as long as their neurological condition permits. Patients should not take naps during the day unless it is absolutely necessary for medical reasons other than MS. When these exceptional circumstances exist, chair rest rather than bed rest should be recommended.

To enable patients to overcome fatigue, maximize energy, and minimize the chances of overexertion we suggest the following: sleep regular hours and avoid unnecessary late nights. Recognize a limit, even though it may vary from day to day. If a prolonged, very tiring task is undertaken, try to do it in steps; work and then rest for a few moments before continuing. Plan the most active hours for early in the day when the energy level is the highest (as detailed in Section 16.4, Planning an Exercise Programme). We find that an organized gradual increase in activities in a structured exercise programme to the limits of the patient's capabilities is most beneficial.

16.3.17 Diet and vitamins

Three diets have received considerable publicity in the past two decades (Crawford, Budowski and Hassam, 1979): the gluten-free diet, the polyunsaturate-supplemented diet, and the low-fat diet. Liversedge (1977), who reported on the consensus of the use of gluten-free diet in 1977, found no indication that the diet had any influence on the course of the disease. Thus, it is highly unlikely that a gluten-free diet is of any benefit in MS.

A stronger case, perhaps, can be made for the polyunsaturate-supplemented diet. This diet was also reviewed by Liversedge (1977). A 2-year

controlled double-blind trial of 75 MS patients revealed that relapses were less severe and of shorter duration in patients given linoleic acid supplementation, but the overall rate of clinical deterioration was not altered (Millar *et al.*, 1973). The dietary supplement caused a substantial rise in the serum unsaturated fatty acid level of patients.

In another trial involving 116 MS patients, also double-blinded and controlled, similar results were obtained. Those patients receiving 23 g of linoleic acid daily and demonstrating increased polyunsaturated fatty acid levels in their blood had shorter and less severe exacerbations. Those patients receiving a much smaller dose of linoleic acid in a different preparation showed no alteration of polyunsaturated fatty acid levels and no clinical effects. However, the rate of clinical deterioration and the frequency of relapses was not altered in either group (Bates *et al.*, 1978).

A third study (Paty *et al.*, 1978) failed to show any alteration in relapse rate or severity despite demonstrated alterations in unsaturated fatty acid levels in the blood.

Finally, Field and Joyce (1979) have proposed that in MS families who have abnormalities in their diagnostic test for MS utilizing polyunsaturated fatty acid levels, MS might be prevented by supplementing the diet of infants with polyunsaturates before the age of 5, the age at which active myelination ends.

Thus, although the relationship of polyunsaturated fatty acids to the possible pathogenesis of MS remains intriguing (Mertin and Meade, 1977), no benefit to established disease by dietary supplementation has thus far been proven.

Swank (1970) has spent many decades following MS patients on a low-fat diet compared to retrospective controls presumably not on his low-fat diet. In spite of drop-outs, he claims the diet is of benefit in decreasing the number and severity of relapses, as well as the total disability.

The low-fat diet proposed by Swank was reviewed critically by Sibley (1970). He stated that it is impossible to say whether or not the low-fat diet is a partially effective therapy, on the basis of all evidence at hand, because an adequate control group of patients consuming an average diet was not studied.

Most megavitamin therapies can be taken without harm. However, there is no evidence that they alter the course of the disease (Poser, 1978).

In summary, many diets and vitamin supplements have been proposed, but none has been scientifically and conclusively proven to be effective in modifying the course or symptoms of MS. We endorse only a well-balanced sound diet approved by scientific nutritionists.

16.3.18 Dorsal column stimulation

Dorsal column stimulation which may be recommended for spasticity, pain and bladder disturbances (Spiegel, 1982) involves the placement of two

16.3.20 Alcoholic beverages

An occasional social drink will do no harm. On the other hand, if the patient suffers from weakness, fatigue, diplopia, dysarthria, dysphagia, or dysbalance, alcohol will probably compound these problems. Most MS patients are not habitual drinkers or abusers of alcohol, so it is easy to abstain or to avoid the second or third drink. Alcohol is dangerous when taken in conjunction with certain medications, and MS patients should abstain under these circumstances.

16.3.21 Informing children

What should the patient tell his/her children about MS? Wasserman (1978) recommends the following:

> 'Depending on their age and maturity, children have varying needs for information and can absorb different amounts. They are quite perceptive and quickly sense when things are wrong.
>
> While they may not need to have the problem identified by name, they do need to be assured, at some level, that their ill parent will not die and leave them alone. They also need to know that even if their parent must take a less active part in physical activities, and even though his or her role in the house may undergo a change, he or she is still able to provide the love expected of a parent.
>
> Children assume that there must be a reason for their parent's illness and frequently conclude that they are to blame. Care should be taken to relieve them of such guilt.'

16.3.22 Travel

Well-planned travel should not be restricted. This is concordant with Bauer's (1977) counsel. The patient should be advised to have a travel companion and to consult a local physician or to phone the primary physician if worsening or complication occurs.

16.3.23 Reading material for MS patients

Many patients wish to read about MS. We recommend that patients join their local MS chapter and their National Multiple Sclerosis Society. The pamphlets and newsletters which each distributes are very informative about coping with MS and about current research advances. In addition, we believe all patients should have a copy of the booklet by Dr H. Bauer entitled: *A Manual in Multiple Sclerosis* (1977) which can be obtained from the National Multiple Sclerosis Society, 205 East 42nd Street, New York 10017, USA.

16.5.1 Disorders of movement

16.5.1(a) Introduction

The information that follows supplements Chapter 20 by Colville entitled 'Rehabilitation'. Disorders of movement in MS result mainly from weakness, spasticity, contractures, dysbalance, incoordination, intention tremor, or combinations of these dysfunctions. In addition, MS dysfunction is made worse by easy or persistent fatigability, disuse, slight rises of body temperature, and/or despair. It is a combination of the above factors, and a natural instinct reinforced by neurologists and family that rest is part of treatment of MS that results in a patient's performing below capacity. To perform below capacity or to be inactive results in disuse and brings on constitutional effects.

A number of complications can occur with disuse in MS: contracture and deformity, negative nitrogen balance contributing to decubitus formation, sepsis and amyloidosis, negative calcium balance contributing to osteoporosis and pathological fractures, renal and bladder calculi, and venous thrombosis with pulmonary emboli (Bauer, 1978). If inactivity can lead to complications, activity should prevent or delay them. It is primarily on this basis that we have emphasized and insisted that MS patients become physically conditioned (see Section 16.4).

Whether exercise is of value in those MS patients who have rapid and continuous functional losses caused by CNS lesions can only be determined by initiating and maintaining a structured exercise programme. If progression is slow, exercise may delay or alleviate disuse and complications, and may even increase function.

The traditional approach to exercise follows from the principle that function begets function. Therefore, if function is the goal, then function is a logical and necessary therapeutic tool. The elements of such an exercise programme could include the following: (1) repetition of individual movements in order to increase strength and endurance; (2) passive stretching of muscles to reduce spasticity and range of motion; (3) exercises utilizing activities of daily living to improve dexterity and coordination; (4) gait training using aids of ambulation (canes, crutches, walkers and braces) as necessary; (5) hydrotherapy to dissipate heat and to utilize buoyancy which permits increased activity and range of motility; (6) use of intact vision to circumvent proprioceptive loss.

Because MS patients have balance problems they are often mistaken for alcoholics. If the balance problem is serious enough to cause a staggering gait the use of a cane or crutches may prevent a catastrophic fall; and, at the same time readily identify the patient as one with a physical disability rather than as a 'drunk'. It is also recommended that the patient wear a bracelet with 'multiple sclerosis' engraved on the back.

Some patients see acceptance of a cane or crutches or wheelchair as a sign

[redacted] to the disease; others view it as a key to greater [redacted] increased independence. Failure to use aids when needed [redacted] participation in social activities and leads to isolation. In [redacted] when medical equipment becomes essential to physical safety, it is foolhardy not to take advantage of what is available.

For those who are wheelchair bound with upper extremity function, a self-propelled vehicle electric motor), such as the Amigo™ wheelchair, is recommended (Amigo™ The Friendly Wheelchair, Amigo Sales, Inc., 6693 Dixie, Bridgeport, Michigan 48722, USA).

A wide range of bathroom safety equipment is available, none of which interferes with the use of the bathroom by others. Such items include elevated toilet seats, grab bars and shower chairs.

There are also numerous devices which make everyday tasks easier: built-up silverware, reachers, page turners, telephone holders, and utility carts. An experienced occupational therapist can make suggestions. Self-help aid catalogues and magazines for the disabled can also be consulted. We recommend a catalogue (1081) by Maddak, Inc. (Peguanmock, New Jersey, 07440, USA) entitled: *There Is A Way*. It displays aids for daily living, home health care and rehabilitation.

16.5.1(b) Weakness

Weakness can be a significant complaint in many MS patients. It is almost always of the upper motor neurone type, and hence is associated with spasticity, clonus and involuntary spasms (McAlpine *et al.*, 1972). In evaluating a patient with weakness, the physician should keep in mind certain aggravating factors that may be present. These include excessive exertion, overuse of muscle relaxants, intercurrent infections, fever, environmental increase in temperature, calcium and electrolyte imbalances, and malnutrition. Loss of strength caused by long-standing MS lesions cannot be reversed by exercise. However, weakness from disuse and despair occurs often and thus a structured programme of physical therapy is recommended (see Section 16.4) to determine if the patient is performing below capacity. Activity to the onset of fatigue is encouraged.

Because the use of braces (especially for foot drop due to heel cord shortening) promotes local atrophy if used prematurely, bracing should be prescribed only when it is felt that ambulation will become safer.

In general, active exercises, especially swimming or jogging, are prescribed for patients with mild to moderate weakness. Extended walks should be encouraged, as well as deep-knee bends on arising and at bedtime. Moderately to severely weak patients who require aids for ambulation or who can transfer to and from the wheelchair and bed patients should be referred to a physical therapist for programmed active and passive exercises. Devices that can be installed in the patient's home to encourage activity and independence include grab bars and hand rails in appropriate places, walker,

parallel bars, a stationary bicycle, and a bed trapeze.

In extreme conditions, a wheelchair or bed existence may be necessary. The physiatrist has many mechanical methods to improve efficiency in these confined quarters (*Multiple Sclerosis: Guidelines for Diagnosis and Management*, 1980).

Frequently, patients with severe paraparesis or paraplegia develop significant oedema. This can be relieved by elevation of the extremity and modest doses of hydrochlorothiazide (Hydrodiuril™).

16.5.1(c) Spasticity

Spasticity, characterized by stiffness of the extremities and hyperreflexia, is one of the most common signs of MS (McAlpine *et al.*, 1972). Patients may describe difficulty in walking or in using the hands in spite of adequate muscle strength. However, it can be considered a useful phenomenon in those patients who have severe paraparesis, since stiff legs can be used to pivot the patent from one location to another. Loss of this pivot function may be a complication of any treatment regimen for spasticity. The patient who has moderate spasticity learns that inactivity will lead to increased stiffness; even an uninterrupted 8-hour sleep leaves the legs stiffer on rising. The patient discovers that arising several times at night (sometimes necessary because of nocturia) and several times during the day (doing 'stand ups') limbers the legs. With patients who have moderate to severe spastic paraparesis, active and passive exercises are most useful. These exercises are also useful for forestalling progressive shortening of muscles. The neutral or slightly stretched position can be maintained by splints, braces, or bivalved cases (Schneitzer, 1978). Massage, hydrotherapy, and the application of external cold can also be useful (*Multiple Sclerosis: Guidelines for Diagnosis and Management*, 1980). A patient with moderate spasticity learns to stand for a few seconds after arising before taking a step in order to achieve balance. She/he limbers up somewhat after the first few steps. The best part of the day is usually between breakfast and lunch, in contrast to modestly ataxic patients, who become more efficient as the day progresses. Spasticity can be aggravated by a number of conditions such as urinary tract infection, decubiti, fever, and rectal or bladder distention.

Excessive spasticity is sometimes a significant barrier to an exercise programme. The energy cost of movement in MS is increased on the contracting muscle because it has to overcome the spastic response in the opposing muscle (antagonist), and even more distant muscle groups stimulated by attempts at movement, which should be relaxing. Antispasticity drugs, such as baclofen (Lioresal™), diazepam (Valium™) and dantrolene sodium (Dantrium™) can be of significant help (Young and Delwaide, 1981a, b).

Baclofen should be initiated with 5 mg twice a day, to be increased by half a tablet every three days to a maximum of two 10 mg tablets four times a day.

16.1.5(e) Balance sense dysfunction

Cerebellar dysfunction may cause difficulty in maintaining an upright posture. Truncal titubation causes swaying from side to side and the patient falls easily. There is no medication or operation for relief of this common difficulty. Frankel exercises are of only limited value. If the dysfunction is associated with weakness, strengthening exercises should be prescribed. The proper use of ambulatory aids such as a weighted cane with a tripod base, Lofstrand crutches, full crutches or a weighted walker can be of great help. Bracing to limit the excursion of the limb to a single plane of movement may be rewarding (Schneitzer, 1978).

The patient learns many ways to compensate for loss of balance sense, such as strengthening the legs by deep-knee bending; pausing after rising for balance before taking a step; broadening the walking base purposely; watching steps; touching or using stair handrails; taking short rest periods after fatigue sets in; and avoiding walking on bumpy surfaces, slippery floors, in crowds, or in the dark. The patient should be encouraged to carry on with vigour, but the gait must be slow and cautious to prevent accidental falls. Exercising in a tub or swimming pool is much easier than on dry land for MS patients because their balance sense is markedly improved in water (see Section 16.4).

16.5.1(f) Incoordination and tremor

A more intractable barrier to the restoration of function is incoordinaton, especially with accompanying tremor. Training may be of some benefit when the condition is mild, but it is of questionable value when severe tremor is present. Fortunately, such a condition brought on by relapse may be relieved by remission.

In the milder stages the patient learns to stabilize movements by reaching with both hands or by bracing the hand while the act is being performed. The tremor often lessens as the day progresses. Excitement and fatigue may provoke it, as will elevation of body temperature. Physiatrists can be of value by reorganizing daily tasks of living and by incorporating a wide variety of assistive devices to accomplish tasks requiring dexterity (e.g. weighted bracelets or weighted eating utensils) (*Multiple Sclerosis: Guidelines for Diagnosis and Management*, 1980). A head rest, of the barber's chair type, or a soft neck collar may minimize head tremor. Attempts to relieve tremor with medication have been unsatisfactory, but diazepam (Valium™), chlorpromazine (Thorazine™), or chlordiazepoxide (Librium™) may be of some value.

In selected patients who have severe intention tremor of both arms, mild to moderate weakness, and insignificant mental retardation, cryothalamotomy has given improvement. The patient should be observed over a 1-year period to determine that the tremor is permanent and progressive prior to the operation (Cooper, 1967). If a patient can scratch his nose, we do not recommend surgery. The symptom may recur after several years; however, the

gratifying results of the initial procedure have encouraged a few patients to ask that it be repeated.

16.5.2 Bladder dysfunction

The information that follows supplements Chapter 17 by Parsons entitled 'The Bladder in Multiple Sclerosis'. Many patients who have MS never experience bladder problems; others are bothered by them only during exacerbation. When they do exist, the inconvenience caused by even minor disturbances as urgency and frequency of urination may interfere with social or business life.

To minimize social limitations by bladder problems we recommend a bladder training programme. This is begun by recording fluid intake and output over a few days to determine how long after drinking the patient needs to void. Next the patient is instructed to schedule regular periods of voiding. An attempt should be made to empty the bladder by relaxing, by thinking emptying of the bladder, by the Crede manoeuvre, or massage of the bladder via the rectum utilizing a finger cot. The amount of time between voiding should be increased to find the maximal interval which comfortably eliminates the risk of incontinence. The bulk of drinking should be done at home or sufficiently ahead of planned outings.

For those cases with little or no residual urine, men find that a condom taped to the penis attached to a tube and leg receptable is helpful. A collection bag strapped to the leg and worn under pants makes the entire apparatus portable and concealable. In females accidents may be avoided by the use of a sanitary napkin or disposable diapers (oversize size), or ABD pads under plastic underpants with reusable liners, all of which are available at pharmacies.

If significant bladder problems (mainly repeated infections and incontinence) exist, a urologist with training in neuro-urology should be consulted. After a thorough evaluation based on the type of neurogenic bladder problem, the correct medications, or mechanical devices, or perhaps surgical intervention can be prescribed to overcome the problem.

A recent review (Blaivas, 1980) of urological problems in MS stresses the poor correlation between symptoms and underlying pathophysiology. All patients should have physiological studies performed at the onset of significant urinary symptomatology. Usually cystometry, urinary flow rate and determination of post void residual volume are adequate. If the diagnosis remains in doubt a cystometrogram and sphincter electromyography should be obtained (Bradley, 1978).

Classification of bladder dysfunction is most practically based on management goals, as recommended by Parsons (Chapter 17). The classes are: (1) failure to store, (2) failure to empty, (3) the combination of both. Failure to store is manifested by frequency, urgency, intermittent hesitation, nocturia, and in its extreme, incontinence. If the patient has moderate

16.5.3 Bowel dysfunction

Abnormality of bowel function in MS can take the form of constipation, incontinence, or frequency and urgency (McAlpine *et al.*, 1972). Constipation is most common in MS and can be due to improper diet, lack of exercise, stress, insufficient rest, irregular habits, MS lesions or a combination (McAlpine *et al.*, 1972). Its treatment and management in mild to moderate manifestation is similar to other types of functional constipation. Regular bowel movement patterns can be achieved in almost every patient by a combination of diet, massage (rectal stimulation), stool softeners, cathartics, and suppositories. We recommend daily bowel movement or one every other day. Increased fluid intake and a diet that includes a fibrous residue (bran) may be adequate for mild cases. If needed a stool softener such as sodium sulphosuccinate (Colace™) 100 mg per day, may be added, with or without a regularly scheduled regimen of oral cathartics and rectal suppositories. The following drugs may be used: milk of magnesia with or without cascara; psyllium hydrophilic mucilloid (Metamucil™); senna concentrate (Senokot™); bisacodyl (Dulcolax™); rectal bisacodyl suppository (10 mg every other day in the morning, which is effective usually within an hour); bisacodyl tablets, 5 mg every other evening, in moderate cases followed by bisacodyl suppository in the morning, and Doxidan™ (a combination of dioctyl calcium sulphosuccinate, a stool softener, and danthron, a mild peristaltic stimulant that acts mainly in the lower bowel). 1 tablet twice a day. Rectal impaction is treated by manual removal as well as enemas. Fleet Enemas™ should be used routinely every other day if necessary (Schneitzer, 1978).

Incontinence and urgency are difficult problems to treat because of the frequent association with paraparesis. The use of anticholinergics may be helpful, or planned evacuation with the use of Ducolax™ tablets in combination with bisacodyl suppositories or Fleet Enemas™ every other day can give relief. In the rare patient whose bowel urgency cannot be managed, colostomy might be indicated for hygenic reasons, especially if sacral pressure ulcers exist.

Rarely do MS patients develop faecal incontinence, unless another condition appears that is associated with diarrhoea. A search for the cause may be appropriate if it persists for more than a few days or is not obviously associated with a viral infection, mild food poisoning, or drugs such as antacids or laxatives. Diphenoxylate hydrochloride with atropine sulphate (Lomotil™) liquid 5 ml four times daily can relieve the acute symptoms. If the condition persists the patient should be evaluated in the hospital.

16.5.4 Visual disturbances

Visual disturbances include oscillopsia and diplopia as well as decreased visual acuity due to errors of refraction, uveitis, cataracts, central scotoma,

field defects and difficulty tracking associated with intranuclear ophthalmoparesis (McAlpine *et al.*, 1972). Patients adjust to these abnormalities by using the direction of gaze that gives the least difficulty. The routine ophthalmological examination should include evaluation for errors of refraction; detection of cataracts frequently associated with steroid therapy (which probably progress less rapidly when the patient is not taking steroids and can be treated with surgical extraction); and a uveitis, which is occasionally associated with MS (McAlpine *et al.*, 1972). The non-infectious uveitis is treated with mydriatics and local corticosteroids. High-powered magnifiers give some help to patients with moderately severe central scotomas or decreased vision secondary to optic atrophy. Patients with field defects and/or difficulty in tracking can improve reading by utilizing an engineer's triangular ruler. If sense of touch is preserved and patients are motivated, the Braille system of reading can be taught. Blind patients with severe ataxia or sensory loss should be made aware of the talking book programmes available through city library systems and local MS chapters (Stetten, 1981). Diplopia often is compensated for in many patients by suppression of one of the images after several months (Schneitzer, 1978). In those cases where it continues to be a problem, a plastic eye shield over one eye and alternated between eyes is recommended. It may also have some value in patients in whom oscillopsia is dissociated.

Acute optic neuritis may occur giving rise to occular pain, impaired visual acuity, and field defects. Prognosis is usually good, with recovery in several weeks. Residual visual loss, however, is common. ACTH or oral steroids have been found to hasten recovery and return vision in the acute situation (Rawson, Liversedge and Goldfarb, 1966).

16.5.5 Dysarthria

The cranial nerves affecting speech mechanism are frequently involved in MS (McAlpine *et al.*, 1972). Speech may become slurred and have low volume. Cerebellar disturbances can cause a dysmetric, scanning speech, and an inability to talk rapidly. Shallow breathing and low breath pressure due to paresis may also affect speech. The MS patient learns that dysarthria may be more severe when she/he is excited, overheated, e.g., owing to weather or fever. Instruction by a professional speech therapist in articulation and breathing can be beneficial, as may progressive resistive exercises given to the muscles of phonation (Schneitzer, 1978). Cerebellar speech disturbances have been less responsive to treatment. Continuous encouragement to speak slowly, distinctly and frequently is important, because the effort to speak can be especially tiring and depressing to some patients with moderate to severe dysarthria. Paroxysmal dysarthria may be treated with carbamazepine (Tegretol™).

16.5.6 Dysphagia

Many patients are aware of mild dysphagia, especially when they drink liquids (McAlpine *et al.*, 1972). Choking intermittently on saliva is also common. Dysphagia can be increased by excitement, tiredness, or being overheated. We recommend gum chewing to strengthen the jaw and swallow mechanism, and to improve coordination.

Rarely is dysphagia a problem, but it often accompanies a fever and then it is one of the most distressing symptoms that the patient can develop. Dysphagia should be considered an emergency under these circumstances because of the potential of pulmonary aspiration. The body temperature should be lowered immediately with a tepid bath or alcohol sponges. Hospitalization should be considered.

16.5.7 Dizziness

Lightheadedness or dizziness, but not true vertigo, occurs in many MS patients with a sudden change of position (McAlpine *et al.*, 1972). This postural vertigo is usually not a bothersome symptom. Attacks resembling acute labyrinthitis manifested by nausea, vomitting, and true vertigo are fortunately of short duration and usually do not recur. Bed rest with restriction of head movement can give relief with or without administration of dimenhydrinate (Dramamine™), meclizine HCL (Antivert™ or Bonine™), chlorpromazine (Thorazine™), or fanityl plus droperidol (Innovar™) (Johnson, Fenton and Evans, 1976).

16.5.8 Paresthesias

There is no effective treatment for paresthesias which are very common in MS (McAlpine *et al.*, 1972). These are often described as 'tingling-picking', 'numbness', 'like after novocain', 'tightness around knees', 'fingers too big', 'can't identify by touching', 'girdle sensation'. Lhermitte's symptom is a possible type of paresthesia. Occasionally analgesics, tranquillizers, or carbamazepine (Tegretol™) may give some benefit.

16.5.9 Trigeminal neuralgia

Trigeminal neuralgia, a specific type of radicular pain, is not a common symptom in MS (McAlpine *et al.*, 1972). When it occurs, the treatment of first choice is carbamazepine (Eadie and Tyrer, 1980). The initial dose of 100 mg three times a day may be rapidly increased over 3 or 4 days to 200 mg three times per day. Beyond this, the dosage should be increased every 7 days to obtain pain relief, if the patient is not unduly troubled by side effects. Doses up to 2000 mg per day may be required, though dosage should be kept as low as possible, consistent with adequate pain relief and adverse side effects. A

blood level of at least 5 μg/ml is necessary and sometimes much higher. Pretreatment and periodic blood counts are recommended because of the potential for aplastic anaemia.

If patients cannot tolerate carbamazepine phenytoin may be used, but it is less effective. A dose of 100 mg three times a day initially should be increased to the patient's limit of tolerance by steps of 100 mg every three days.

Clonazepam has also been shown to be effective. An initial dose of 0.5 mg twice a day may be increased at intervals of 5 days. When the pain is controlled the dosage should be kept as low as possible and tapered at three months to determine if it is still needed. Baclofen also has been reported to be effective in a dose of 30–80 mg day^{-1} in three or four divided doses. Some patients were unable to tolerate this dosage schedule because of drowsiness or gastric distress (Fromm, Terrence and Chattha, 1980). If the pain proves refractory to drug treatment combinations, percutaneous radiofrequency stereotactic thermal rhizotomy of the appropriate trigeminal nerve branch is recommended (Sweet and Wepsic, 1974; Tew and Keller, 1977; Dalessio, 1981).

16.5.10 Pain

Pain due to MS lesions is not a common feature of MS in our experience or that of McAlpine *et al.* (1972). On the other hand, in another study 42% of MS patients had some form of pain (Carer, Sciana and Merritt, 1950). Pain can take many forms, as for example headache, backache, neckache, muscle cramps and spasms, and dysalgesia (Bauer, 1977). Many MS patients have a chronic backache and neckache that is probably secondary to spasticity or strain on the low back area produced by normal gait or posture. Mild analgesics (aspirin or acetaminophen), muscle relaxants (baclofen, diazepem, methocarbamol (Lobaxin™) or carisoprodol (Soma™), cyclobenzaprine (Flexeril™)), physical therapy, bed boards, local heat, and hydrotherapy may be helpful. During an acute flare-up of backache bed rest on a firm mattress with leg elevation may be necessary. Muscle cramps trouble only certain patients, and muscle relaxants, baclofen and diazepam, can give significant relief.

Pseudoradicular pain, probably due to involvement of the sensory root zone or spinothalamic tracts of the spinal cord, can be particularly troublesome because of its refractoriness to the milder analgesics. It is usually manifested by a burning sensation, predominantly in the lower extremities. Opiates or codeine except in the acute situation should be avoided to prevent dependency. Phenytoin (Dilantin™) and carbamazepine (Tegretol™) have been effective for control of this pain in some cases. Pretreatment and periodic blood counts are recommended for patients receiving carbamazepine because of the possibility of bone marrow suppression. Phenol nerve root blocks, posterior root section, or cordectomy may be considered in rare cases refractory to other therapy.

16.5.11 Convulsions

The incidence of convulsions is higher in patients with MS than in the general populace. In one recent study (Gozzoli, Faggi and Cosi, 1979) 10% of clinically definite MS patients had seizures sometime during the course of the disease. The investigators compared this with other reports where the average percentage was 5.2% with a range of 2.2–15%. On the other hand, Ritter and Poser (1974) did not find an increase in MS. Therefore, a search for other causes of convulsions must be made. All types of seizures have been seen, but the most common are generalized and local motor seizures. Only petit mal epilepsy seems to be excluded. In many instances different types of seizure occur in the same patient (Gozzoli *et al.*, 1979). Excellent relief can usually be obtained with phenytoin (Dilantin™), 100 mg three times a day or the usual anticonvulsion doses of carbamazepine (Tegretol™). After approximately 6 months of a convulsion-free period, the anticonvulsant dose can be tapered and finally discontinued over a 3-month period with treatment restarted if the symptom returns.

16.5.12 Tonic seizures

Tonic seizures in MS have been reviewed by Matthews (1975) and Twomey and Espir (1980). The incidence is as high as 17% in the Japanese literature. Above half of the patients note precipitating factors, the majority being triggered by tactile stimulation or trunkal movements. The limbs go into spasm without associated movements or loss of consciousness. The spasms are painful in about half of the cases reported. The pathophysiology of tonic seizures remains conjectural. A possible explanation is that impulses, especially afferent ones, reach an area of demyelination and spread laterally to neighbouring afferent and efferent axons within the plaque of demyelination. The response of all types of paroxysms in MS (tonic seizures, paroxysmal dysarthria, ataxia, diplopia, paresthesias, pains, itch and akinesia) to Tegretol™ is often dramatic.

16.5.13 Dementia

Impaired intellectual functioning varies widely among recorded studies, ranging from 2 to 72% (McAlpine *et al.*, 1972). In one recent report, 64 patients, all with severe longstanding disability ranging from 3 to 39 years, were given a battery of tests to measure intelligence; 64% had some evidence of memory impairment (Staples and Lincoln, 1979), but less than 10% had severe dementia and this was associated with severe disability. The abilities most affected are memory and abstract reasoning. When modest to severe dementia is present for which there is no treatment, it interferes with rehabilitation, and custodial care may be necessary.

this section is entitled PUTATIVE TREATMENTS. i.e. treatments aimed at proposed specific disease processes.

Poser (1978) stated:

> 'The Herculean aspects of evaluating the efficacy of any form of treatment can best be exemplified by the pessimistic attitude of a number of neurologists who stated 35 years ago that no treatment can be properly evaluated because of the confusion caused by remissions, and, for this reason, any therapeutic endeavor is futile.'

We believe this pessimistic attitude has been reversed by a report by Brown *et al.* (1979) which isolated the problems of assessing specific therapies for MS: (1) Precise diagnostic criteria are necessary. (2) Due to the variability of the disease in each patient, evaluation of any form of therapy is extremely difficult, and any new approach must be treated with some scepticism until results can be confirmed by others. (3) It is necessary that standardized disability scales based on qualitative and quantitative measurements be used to follow patients. (4) Studies to investigate the clinical usefulness of a particular therapy must have large groups of patients followed for a 'sufficiently long time' and comparisons made with a concurrent sizable control group. A double-blind design utilizing standardized qualitative and quantitative clinical measurements, CNS IgG synthesis, evoked potentials, and relevant check lists to record relevant clinical side effects and laboratory adversities must be used. (5) A biostatistician should be employed in all aspects of the experiment.

The report by Brown *et al.* (1979) concluded that a well-designed MS clinical trial may well develop useful scientific information even if the putative therapy fails to show any therapeutic effect. Therefore, based on that report, we have in hand a reliable and valued set of guidelines to carry out clinical trials in MS.

Four other advances have been made in the last decade which we believe further strengthen the point of view proposed by Brown *et al.* (1979): (1) Standardization of serial clinical quantitation of neurological function (Potvin *et al.*, 1980). Now the objective quantitative data, obtained by a trained technician who is unaware of treatments or adversities, can be calculated in terms of percentage of normal function during the course of a clinical trial (time vs treatment vs a given neurological function (e.g. strength)) (Henderson *et al.*, 1978). (2) Application of evoked potentials, especially visual evoked potentials, to detect subclinical lesions with or without hyperthermia (McDonald, 1980; Potvin *et al.*, 1980; Sal and Selhorst, 1981). Objective electrophysiological data obtained by a trained technician who is unaware of treatments or adversities can be calculated in terms of percentage of normal function during the course of a clinical trial. (3) Delayed super-enhanced CT brain scans to detect *in situ* changes in the MS blood–brain barrier (McCammon *et al.*, 1981). The use of a method to evaluate neuropathology of

that these regimens were any better than some reports for single agents, and progression of disease continued despite their use in combination.

Mertin *et al.* (1980) reported preliminary results of a continuing double-blind controlled trial of immunosuppressive treatment of MS patients with a combination of antilymphocyte globulin, prednisolone and azathioprine. In the first 30 patients completing the 15-month treatment period there appeared to be a reduction in the number of relapses and some retardation of the clinical course of the disease in the immunosuppressed group compared to the placebo group ($P<0.05$). The beneficial effect of the treatment was only seen in females.

Marforio *et al.* (1981) did a long-term study utilizing corticosteroids or corticosteroids and azathioprine. The course of the disease was less severe in patients treated with the combination.

16.7.4(i) Conclusions

From the above it can be concluded that the frequency of relapse, rate of progression, and perhaps CNS IgG synthesis can be decreased with the use of immunosuppressive agents, although the few controlled studies show less benefit than those that are uncontrolled. Disease progression is not halted, however, and any effect these agents have is probably temporary. Many researchers report eventual acceleration of progression back to control levels once therapy is discontinued. Patients have not been followed long enough in most cases to evaluate delayed side effects such as carcinoma. Patients early in their course may be most benefitted, but most have mild disability by this time. Thus, any temporary benefit must be weighed against possible long-term serious adverse effects, which have not been adequately assessed. For the present, routine clinical use of these agents is not warranted, and their use should continue to be confined to planned research.

16.7.5 Other modalities

16.7.5(a) Myelin basic protein

Myelin basic protein was proposed as a possible therapeutic agent for MS when it was discovered that it could suppress the induction of EAE (Romine and Salk, Chapter 19). Three studies of myelin basic protein administration to MS patients have brought negative results (Campbell *et al.*, 1973; Gonsette, Delmotte and Demonty, 1977; Romine and Salk, Chapter 19).

Alvord *et al.* (1979) questions whether myelin basic protein has received a fair trial in the treatment of MS. In their studies with monkeys, a non-specific adjunctive factor, an antibiotic and a corticosteroid were also required. Accordingly, they believe that human trials of the therapeutic efficacy of myelin basic protein in MS should include the administration of large doses of myelin basic protein together with adjunctive agents.

study by Gonsette *et al.* (1981) indicated that neurological function and disability status deteriorated over a 2-year period, whereas the levamisole-treated groups remained stable. No serious adversities were noted. They concluded that levamisole could be beneficial to patients with a progressive type of course.

16.7.5(e) Plasmapheresis

The rationale for the use of plasmapheresis in MS patients is commented on by Dau *et al.* (1980). They suggest that removal of immune complexes by plasmapheresis may be beneficial to MS patients since complement is preferentially localized to the peripheral zone of active demyelination in plaques along with IgG. The preliminary study by Dau *et al.* showed modest improvement in neurological function for 7 of 8 patients classified as progressive MS and subjected to long-term plasmapheresis in combination with azathioprine and pulsed prednisone therapy. In 6 of 7 patients CSF IgG decreased. They recommended that a controlled trial was necessary to confirm these preliminary results. Could it be that the improvement found in this experiment was the result of the prednisone therapy and not the plasmapheresis and azathioprine?

Along this line, Tindall (1981) did a prospective randomized trial of plasmapheresis and azathioprine administration. Based on the rationale that MS is an immunologically mediated disorder in which demyelinating factors have been reported in serum and CSF and that plasmapheresis has been reported to improve MS in preliminary studies, a prospective, randomized study of plasmapheresis and azatheioprine vs azathioprine alone was carried out. Twenty patients with chronic progressive MS and progression in the previous 12 months were randomized to either 3 mg kg^{-1} daily of azathioprine for 12 months, or to azathioprine and plasmapheresis. Three 5-litre exchanges were performed in 7 days, followed by one exchange every 3 to 4 weeks for 12 months. This format maintained circulating immunoglobulins below normal for the duration of the study. Disability was evaluated at entry, 6 months, 1 year, and exit. Subjective improvement occurred in the plasmapheresis group during the first 1–2 weeks, but without objective alteration in the disability scale, and was not sustained. No significant improvement was seen in either group. Neither plasmapheresis plus azathioprine nor azathioprine were effective in producing objective clinical improvement or in halting progression.

Khatri *et al.* (1980, 1981) studied 24 consecutive patients with chronic progressive MS utilizing weekly plasmapheresis in conjunction with daily cyclophosphamide and alternate day prednisone. All patients improved. Normalization of suppressor T-cell function correlated with clinical improvement.

Lymphocytopheresis was carried out by Giordano *et al.* (1980 on 50 patients with progressive MS. Ninety-four per cent demonstrated subjective

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CHAPTER NINETEEN

A Study of Myelin Basic Protein as a Therapeutic Probe in Patients with Multiple Sclerosis

John S. Romine and Jonas Salk

19.1 Patients
19.2 Cinical procedures
19.3 MBP and course of administration
19.4 Course of disease in MBP and placebo-treated patients
19.5 Side effects
19.6 Discussion

Experimental allergic encephalomyelitis (EAE) has been studied intensively by many investigators as a presumptive model for multiple sclerosis (MS) (Adams, 1959; Alvord, 1965; Patterson, 1976; Raine, Traugott and Stone, 1978). In the course of investigations with this model, it has been found that EAE in experimental animals can be prevented or suppressed by daily subcutaneous administration of large doses of purified heterologous myelin basic protein (MBP) (Salk *et al.*, 1980). Since this could be done without harm in animals, it was of interest to carry out similar studies in MS patients. The use of MBP, as a therapeutic probe in this way, was intended to reveal whether or not EAE of known cause and MS of unknown cause have similar responses to MBP.

As will be revealed in the details presented in this chapter, the uniformity and character of the therapeutic response of MBP in EAE is not evident in patients with MS; a suggestive beneficial effect of uncertain significance has been seen in a few and no therapeutic effect has been observed in the majority of patients. These observations indicate that the aetiology of MS is different and more complex than that of EAE and also suggest the possibility that MBP may have a beneficial effect in a sub-population of MS patients.

Further studies will be required to confirm the suspicion that MBP may have a favourable effect in a sub-group of MS patients. Confirmatory studies have been slowed by the occurrence of delayed-type hypersensitivity reactions at injection sites, and attempts are presently being made to overcome this effect. If successful, then larger-scale studies can be carried out.

Immunological observations made thus far have revealed the presence of

were the changes in neurological function dramatic or life-threatening. In six patients, recovery to the baseline level of function was prompt and complete following interruption or discontinuation of injections and, in some instances, after a course of prednisone. In two patients, recovery was short of baseline and the residual deficit was mild in one and moderate in the other.

Table 19.5 Hypersensitivity side-effects of MBP in MS patients

	Frequency
Delayed-type at injection site	15/19
No neurological changes	7/15
With neurological changes	8/5
Reversible	6/8
Residual	2/8
Immediate-type generalized	2/19

Continued administration of MBP at dosage levels of approximately 1–3 mg kg^{-1} day^{-1} has been well tolerated for up to two years without evidence of adverse clinical or immunopathogenic effects. A variety of methods have been tried to avoid or ameliorate the early delayed-type reactions including pretreatment with prednisone and modification of the initial regimen of MBP. So far, none of these approaches have been successful. For the prevention of these reactions, prior administration of MBP intravenously at weekly intervals for 2–3 weeks before daily subcutaneous administration of MBP is presently under investigaton. The rationale for this approach derives from the work of Claman (1976) on the induction of tolerance to a delayed hypersensitivity inducing antigen in mice.

A mild generalized immediate-type allergic reaction occurred on a single occasion in each of two patients instantly following subcutaneous injection after several months of daily administration of MBP. The symptoms were similar in both instances: lightheadedness, a sensation of flushing, a feeling of tightness in the chest, and a transient urticarial rash.

19.6 DISCUSSION

MBP was tested by earlier investigators as a therapy for MS (Campbell *et al.*, 1973; Gonsette, Demotte and Demonty, 1977). Relatively small doses (5.0 mg per dose) were given intramuscularly at weekly intervals; no clearcut benefit or harmful effects were observed in either study. However, an equivalent dosage and regimen of MBP would not suppress EAE in animals and, if EAE is believed to be a model for MS, would not be expected to have an effect on the course of MS (Alvord *et al.*, 1979; Salk *et al.*, 1980). Accordingly, the

dosages used in the present study are larger than any previously used in humans.

Unless MBP is tested in MS patients under conditions known to be effective in EAE, the results of such a test would be inconclusive. In the present study, MBP was administered to MS patients according to a regimen known to be effective in suppressing EAE in animals. The failure to reproduce in MS patients the effects observed in EAE in animals suggests that the two diseases are aetiologically distinct and that, at most, EAE is an incomplete model for MS.

In view of the foregoing, the observation suggesting improvement in two of the patients was unexpected and remains as yet unexplained, especially in view of the continued presence of an abnormally elevated rate of IgG synthesis in the CNS even during the period of neurological improvement (Salk *et al.*, 1980). It will be of interest, therefore, first to determine if the unexpected findings are repeatable, and if so, then to look for a mode of action through an effect on immunoregulatory factors.

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