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ORIGINAL ARTICLE

Multiple Sclerosis: Microglia, Monocytes, and Macrophage-Mediated Demyelination

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Abstract

This study examined the roles of microglia and monocytes in myelin destruction in patients with early multiple sclerosis (MS). Twenty-two cases were studied; the clinical duration was <9 weeks in 10 cases. Twenty myeloid cell subtypes or categories were identified including 2 cell types not known previously to occur in demyelinating diseases. Commencing myelin breakdown in plaques and in perivascular and subpial tissues occurred in the immediate presence of infiltrating monocytes and was effected by a homogeneous population of IgG-positive Fc receptor-bearing early phagocytes interacting with abnormal myelin. Oligodendrocyte apoptosis was observed in intact myelinated tissue bordering areas of active demyelination. Capillaries in the cerebral cortex plugged by large numbers of monocytes were common in acute cases of MS and in a patient with a neuromyelitis optica variant and extreme systemic recruitment of monocytes. In an MS patient with progressive disease, microglial nodules centered on MHC-II-positive capillaries plugged by monocytes were present in the cerebral cortex. This constitutes a new gray matter lesion in MS.

Key Words: Acute multiple sclerosis, Antibody-dependent cellular phagocytosis, Oligodendrocyte apoptosis, Secondary progressive MS, Monocyte encephalopathy, Myeloid cells, Subpial demyelination.

INTRODUCTION

Charcot and other early investigators, using stained sections and teased wet specimens of chronic plaques, noted that affected tissue consisted of packed glial fibers and nerve fibers without myelin sheaths (1) (Supplementary Data Fig. S1). Towards the edges of the lesions there were lipid-filled "fat granule cells," which were thought to be a form of cell death caused by lipid material accumulating within aging or diseased cells. Myelin loss with preservation of axons was attributed to, "strangulation" of myelinated nerve fibers by increasing glial fiber formation.

J.W. Dawson, in a remarkable 1916 monograph (with 155 references, 22 drawings, and 434 photographs and photomicrographs), written about 40 years after Charcot, summarized the work of French and German pathologists and results of his own study of 9 multiple sclerosis (MS) cases (2). He noted that fiber formation was minimal or absent in early lesions and that the first and most important structural change was a commencing loss of myelin with relative preservation of axons, concluding that ". . . changes in the myelin sheath must be looked upon as the primary structural element attacked by. . . some noxious agent or effect . . . from the blood or perivascular spaces." His suggestion as to the nature of the "causal agent" was that it could be a toxin, some unknown pathogen, or an enzyme. How fat granule cells, that is lipid macrophages, came to contain myelin breakdown products he suggested was that myelin in a soluble form was adsorbed by the cell. His illustration of the earliest structural change in newly forming lesions is a colored drawing of several small, lipid-filled fat granule cells located in otherwise normal looking myelinated tissue.

Macrophage-Mediated Demyelination

Waksman showed that destruction of myelin in the T cell mediated autoimmune disease experimental allergic neuritis (EAN), which was produced by injecting intradermally peripheral nerve myelin in susceptible animal strains, occurred near blood vessels and in the immediate presence of blood born leukocytes, mostly macrophages together with other inflammatory cells (3). Later, electron microscopical studies showed that myelin breakdown in EAN and in experimental autoimmune encephalomyelitis (EAE), a T-cell-mediated autoimmune disease of CNS myelin, was effected by macrophages directly contacting myelin sheaths, that they removed myelin directly from myelin sheaths by phagocytosis, and that this occurred without the formation of extracellular myelin debris or engagement with other cells or membranes in the vicinity (4, 5). Referred to as macrophage-mediated demyelination, this pattern of myelin destruction has since been described in other PNS and CNS demyelinating diseases and experimental models including chronic inflammatory demyelinating poly-

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neuropathy, several caused by viruses, and, in the PNS, by localized trauma (6–8).

Regarding MS, of the more than 70 electron microscopy (EM) studies published since 1964, in the few instances where suitably fixed early lesions have been available, commencing loss of myelin has followed this pattern with the additional detail that detached myelin lamellae are sometimes seen attached to receptor-rich areas on the macrophage surface (clathrin-coated pits) (9, 10). A subsequent study has shown the same pattern of macrophage myelin engagement in EAE (11). There are also reports that myelin engaged by macrophages in MS may appear vesiculated and that this may involve the full thickness of the sheath (12).

As to the origin of the macrophage population in MS and other conditions where myelin disruption occurs, the traditional view has been that in all acute forms of injury to white matter including traumatic, ischemic, and inflammatory lesions, lipid macrophages develop from a population of small unusually shaped cells with fine branching processes discovered and named microglia by del Rio Hortega (13). Ramon y Cajal (14) and most modern neuropathologists, while acknowledging the importance of Hortega's discoveries, consider that macrophages including lipid macrophages derive in part or entirely from monocytes from the blood (15–18).

Oligodendrocyte Loss

Oligodendrocytes, sometimes in relatively large numbers, are present in typical postphagocytic plaques (19). The numbers, however, are less than in normal tissue which has encouraged the view that macrophage myelin engagement in MS may be due not to antibody or T cells targeting myelin but to macrophages responding via innate immune mechanisms to the presence of degenerate myelin secondary to oligodendrocyte loss. There are 2 contrasting current opinions regarding this possibility. Brück et al (20) examined oligodendrocytes in series of MS cases diagnosed by needle biopsy of single lesions in cases of clinical duration 11 days to 7.5 months. Some lesions showed oligodendrocytes largely preserved whereas in others oligodendrocyte loss was pronounced. In other series, it was determined that about one third of cases had lesions with oligodendrocyte loss, whereas in two thirds of cases biopsied lesions showed no loss of oligodendrocytes (21–23). Myelin destruction in the latter group was ascribed to the presence of antimyelin antibodies as indicated by the presence of activated complement and IgG on affected sheaths.

The other view, based on autopsy studies of multiple lesions in equally atypical cases of early MS, is that in all cases of MS there is a complete or almost complete loss of oligodendrocytes in newly forming lesions, that the relatively numerous oligodendrocytes sometimes seen are newly generated cells, and that deposition of activated complement (but not IgG) on myelin is common in early lesions in early MS (24–28). Others report no evidence of patient heterogeneity with respect to the presence of myelin immunoreactive for activated complement in patients with chronic disease (29).

Regarding the relative roles played by microglia and monocytes in myelin destruction in MS, some authors ascribe such activity to microglia, others noting that myelin phagocytes are mostly derived from activated monocytes (30–37). This study focuses chiefly on the different morphologically defined populations of microglia, monocytes, and other types of myeloid cells involved in initiating tissue breakdown and myelin destruction in newly forming lesions in patients with early MS.

MATERIALS AND METHODS

This is the second of 2 studies designed to investigate the pathogenesis of plaque formation in exceptionally early cases of MS. The first of the 2 studies reported that astrocytes as well as oligodendrocytes and myelin are destroyed by macrophages in newly forming lesions (38). In this study of the same group of patients, attention is focused specifically on the role played by microglia and monocytes in the destruction of myelin in newly forming lesions.

Clinical Material

The neuropathologists and institutions that provided tissue together with clinical details of 12 acute cases and 10 chronic cases are shown in Table 1. Identifying case numbers are the same as those used in the astrocyte study (38). The study complies with the requirements of the Human Ethics Committee of The University of Sydney.

Immunohistochemistry

Areas of different histological age within lesions are defined as follows: myelinated prephagocytic areas, active demyelination (Luxol fast blue [LFB]-positive macrophages in partially demyelinated tissue), "immediate" postphagocytic zones (LFB-positive macrophages in completely demyelinated tissue), postphagocytic zones (LFB-negative lipid macrophages in demyelinated tissue), and "late" post-phagocytic areas (lipid macrophages numerous but located chiefly in perivascular spaces). Previously identified indications of imminent myelin breakdown in normally myelinated tissue bordering areas of active demyelination (prephagocytic changes), include chiefly loss or degenerative changes in oligodendrocytes, intramyelinic edema, and myelin sheaths immunoreactive for activated complement.

Paraffin sections 4–6 microns thick were stained using hematoxylin and eosin, LFB-PAS for myelin, and Bodian silver stain for axons. Frozen sections were prepared from mirror blocks of selected lesions and stained for neutral lipids using Oil Red O and hematoxylin. Immunohistochemical staining was performed as described in Supplementary Material using primary antibodies listed in Table 2, biotinylated or polymer-bound horseradish peroxidase-labeled second antibodies (Vector ABC Elite Kit, Vector Laboratories, Burlingame, CA; EnVision+ and LSAB+ Kits, Dako Cytomation, Inc., Carpinteria, CA), and diaminobenzidine as chromogen. Antigens and lectins examined included: RCA-1 lectin (endothelial cells, microglia, macrophages), MRP-14 (activation monocyte marker), HAM56 (macrophages), CD45 (myeloid cells), CD209 (dendritic cells), MHC-class II antigens, UCHL1 (CD45RO activated and memory T cells), lymphocytes (CD3, CD4, CD8, and CD20), IgG, complement proteins (C3d, monoclonal B7 anti-MAC 5b-9), and PCNA and Ki67 (proliferating cells). Also examined were sections immunostained using biotinylated normal and MS CSF IgG as described in Supplementary Material as well as sections pre-

TABLE 1. Multiple Sclerosis Cases

Case	Sex/age	Duration of Illness/Terminal Illness	Neuropathologist	Institution
1	F 31	3 months/7 days	CWM Adams	Guy's Hospital Medical School, London, UK
2	F 20	2 weeks	CWM Adams	Guy's Hospital Medical School
3	M 25	29 days	S Pogacar	Brown University Medical School, Providence, RI, USA
4	F 42	2.5 months/2 weeks	RD Terry	Albert Einstein College of Medicine, The Bronx, NY, USA
5	M 14	18 days	J McLaughlin	Royal Free Hospital, Hampstead, London, UK
6	F 70	21 days	S Love	The National Hospital, Queen Square, London, UK
7	M 32	10/8 months	R Doshi	The Maudsley Institute of Psychiatry, London, UK
8	F 48	18 days	CG Harper	Royal Perth Hospital, Perth, WA, Australia
9	F 14	9 months/17 hours	GN Budzilovich	New York University Medical Center, NY, USA
10	M 36	3 years/not known	DM Boehme	VA Hospital, East Orange, NJ, USA
11	F 23	5 weeks	CJ Bruton	Runwell Hospital, Wickford, UK
12	F 23	30 days/60 hours	BA Kakulas	Royal Perth Hospital
13	F 27	32 months	RO Barnard	Maida Vale Hospital for Nervous Diseases, London, UK
14	F 29	12 months	W Evans	Oliver Latham Laboratory, Macquarie Hospital North Ryde, NSW, Australia
15	F 27	49 months	TH Moss	Frenchay Hospital, Bristol, UK
16	M 34	3 years 11 months	DM Boehme	VA Hospital, East Orange, NJ, USA
17	M 43	13 years	JW Prineas	Dover General Hospital, NJ, USA
18	M 55	"Long standing"	CJ Bruton	Runwell Hospital
19	F 39	14 years	JW Prineas	Concord Hospital, Sydney, Australia
20	F 30	11 years	CS Raine	Albert Einstein College of Medicine
21	M 60	20 years	Eun-Sook Cho	University Hospital, UMDNJ—New Jersey Medical School, Newark, NJ, USA
22	M 55	20 years	Dr Krumerman	Jersey Shore Medical Center, Neptune, NJ, USA

Remyelinating lesions were present in the same section or elsewhere in cases 1, 3, 4, 9, and 10. Cases 3, 7, 9, and 10 received corticosteroids. Case 3 received azathioprine. Prominent concentric bands were present in Case 8. Neurogenic pulmonary edema was present in cases 9 and 10. Herniation was present in cases and 8. Cases 16 and 17 received cyclophosphamide. Cases 1–12 are acute cases; cases 13–22 are less acute cases.

TABLE 2. Antibodies

Antigen	Clone	Dilution	Antigen Retrieval	Source
CD45	PD7-26,2B11	1:200	Microwave/citrate	Dako, Carpinteria, CA
CD68	PG/M1	1:50	Pronase	Dako
MAC-1	HAM56	1:1000	Proteinase K	Dako
MHC-class II	CR3/43	1:50	Heat/high pH Dako	Dako
MRP14	BMA-S36,48	1:25	Microwave/citrate	Accurate Chemical, Westbury, NY
RCA-1 lectin		1:4000		Vector, Burlingame, CA
CD209 DC-SIGN		1:100	Heat/high pH Dako	R and D Systems, Minneapolis, MN
IgG		1:8000	Proteinase K	Dako
C9neo (MAC)	B7	1:20	Proteinase K	BP Morgan, Cardiff University, Cardiff, UK
CD3		1:50	Microwave/citrate	Dako
CD4		1:10	Microwave/citrate	Novocastra, Newcastle Upon Tyne, UK
CD8	C8/144B	1:100	Heat/high pH Dako	Dako
CD45RO	UCHL1	1:150	Microwave/citrate	Novocastra
PCNA	PC10	1:80		Dako

pared during previous immunohistochemical studies of the same lesions. The latter included sections reacted for myelin basic protein (MBP), myelin oligodendrocyte glycoprotein (MOG), CNPase, HNK-1 (immature oligodendrocytes and type 2 astrocytes), activated caspase-3, glial fibrillary acidic protein, and aquaporin 4 (AQP4).

Electron Microscopy

Micrographs from previous EM studies of plaques in Cases 19 and 20 were reviewed. In Case 19, the plaques examined were inactive lesions with few or no lipid macrophages. In Case 20, the lesion examined was an actively demyelinating plaque described previously by Raine et al (19).

TABLE 3. Immunohistochemical Profiles of Morphologically Defined Myeloid Cells in and Near Multiple Sclerosis Lesions

Antigen	Ramified	Early Activated	Phagocytic	Lipid	Residual	Monocytes	Plugs
CD45	+	+	+	0 – ±		+	+
CD68	± – +	+	+				
HAM56	0 – ±	+	+				
RIC1	0 – +	+	+				
MHC-II	0 – ±	+	+	±		+	+
MRP14	0	(+)	0 – +	0 – ±		+	+
CD209	0	0	0	0		0	0
IgG	0	0	+	0 – ±		+	+

Ramified: resident and reactive microglia with thin branching processes. Early activated: irregularly shaped or elongated thin myeloid cells with no or equivocal branches. Phagocytic: cells containing particles of Luxol fast blue (LFB)-positive myelin. Lipid: LFB-negative cells distended with lipids. Residual: parenchymal phagocytes with distinctive secondary lysosomes, which are common in chronic multiple sclerosis cases. Currently there are no reliable immunohistochemical data on this population. Monocytes: typical appearance by light and electron microscopy (negative for all lymphocyte markers tested). Plugs: squeezed monocytes in gray matter capillaries.

Blank boxes indicate information lacking or uncertain. + = positive, ± = variable, 0 = negative, and (+) = Case 7.

RESULTS

Myeloid Cells

Resident Ramified Microglia

These nonphagocytic (no phagosomes detected by light microscopy), IgG-negative, MHC-II-negative, small, ramified microglia in normal tissue remote from plaques appeared as depicted by del Rio Hortega and others in drawings and photomicrographs of normal and diseased tissue using particular silver impregnation techniques. Immunohistochemically, these CD45-positive cells stained negatively for all lymphocyte markers tested and they were negative for CD209 and the activation marker MRP-14 (Table 3). In some sections, ramified microglia stained positively for CD68. Ramified microglia were seen encircling (corralling) cell bodies of neurons and other cells in intact gray matter and white matter in several of the acute cases (Fig. 1).

Reactive Ramified Microglia

These enlarged ramified cells were present in increased numbers (microgliosis) near and distant from lesions. They

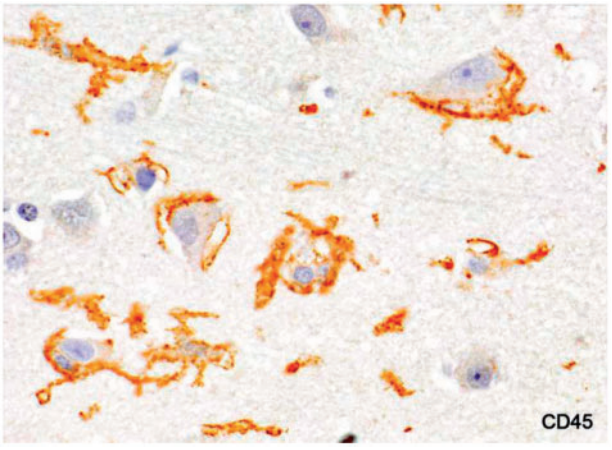

FIGURE 1. Microglia corralling neurons in intact gray matter (cerebral cortex) (Case 7, CD45, ×280).

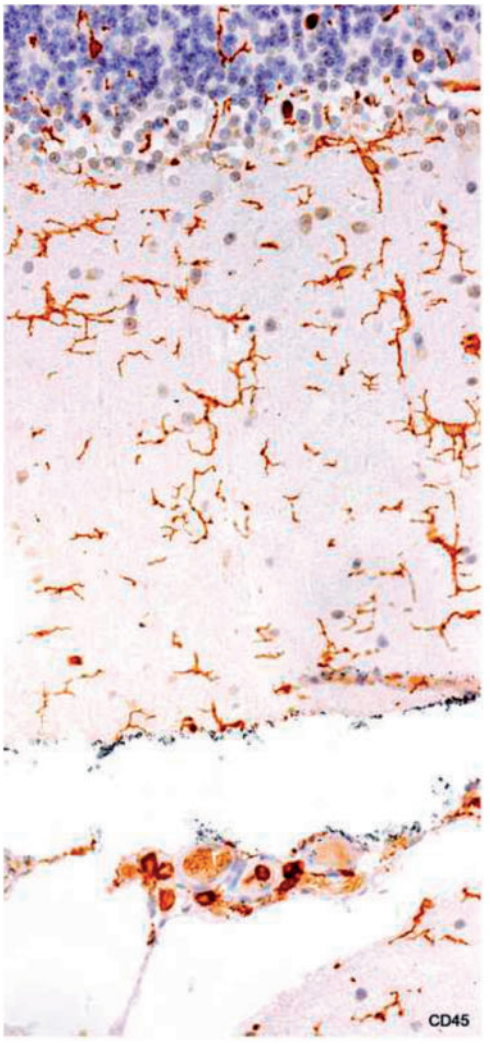

FIGURE 2. Reactive microglia. Enlarged ramified microglia in the molecular layer of the cerebellum. A blood vessel located in the subarachnoid space is cuffed by monocytes (Case 17, CD45, ×220).

were nonphagocytic (no phagosomes), IgG-negative, and MHC-II-negative (Table 3; Figs. 2 and 3). There was a single instance of ramified microglia that were MHC-II-positive and corralling unidentified cells in tissue bordering an early expanding lesion in Case 4 (Fig. 4).

Early Activated Microglia

These elongated cells, irregular in shape or thin, with no or equivocal branches were not phagocytic (no phagosomes by light microscopy), IgG-negative, variably MHC-II-positive, and located in intact tissue bordering lesions (Table 3). They were present in large numbers and were distributed diffusely or in the form of small clusters of IgG-negative microglia (microglial nodules). Although not proven, there was the impression that the blood brain barrier in such areas was intact, (that is no IgG in astrocytes or elsewhere; Fig. 5, Supplementary Data Figs. S3 and S4). This population corresponds roughly to Hortega's nonphagocytic "rod cells" and "flat cells" (13). In the absence of monocytes, myelin and oligodendrocytes in the vicinity usually appeared normal with no evidence of commencing myelin breakdown.

Nodules of Early Activated Microglia Centering on C3d-Positive Myelin

Microglia clustered alongside myelin sheaths immunoreactive for activated complement (C3d), were located in intact tissue bordering lesions (Fig. 6). Such nodules were observed in 2 patients with secondary progressive MS (Cases 16 and 17). The cells were MHC-II-positive, CD16-positive, MRP-14-negative, and IgG-negative. The nodules were seen only in myelinated tissue, never in demyelinated tissue. This and reports that the C3d immunoreactivity may be nodal or paranodal, that paranodes at plaque margins show immunohistochemical evidence of extensive damage as well as EM evidence of detachment of lateral loops from the axolemma, support other evidence that a hypothetical MS antigen may prove to be a nodal/paranodal protein (28, 39–41).

Nodules Composed of Microglia and Monocytes

These were common in intact tissue bordering some lesions, especially in Cases 7 and 17. These nodules were not seen in demyelinated tissue (Supplementary Data Fig. S2).

Nodules Composed of Monocytes, Early Activated Microglia, and MHC-II-Positive Capillaries

Nodules with these characteristics were present in moderate to large numbers in the cerebral cortex in a case of secondary progressive MS (Case 16; Fig. 7). Microglia within each nodule appeared to be encircling (corralling) an MHC-class II-positive capillary containing a monocyte (Figs. 8 and 9). Neurons in the vicinity were intact but showed increased immunostaining of neurofilaments. As described below,

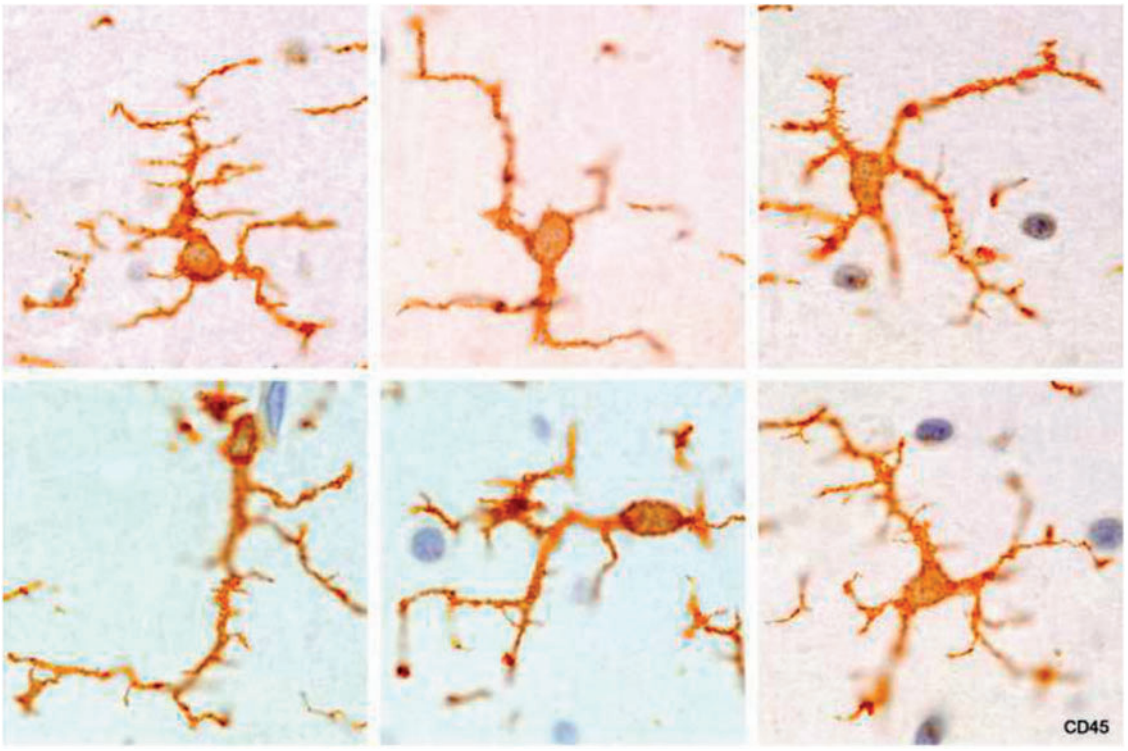

FIGURE 3. Reactive microglia in the molecular layer of the cerebellum. Cell bodies consist of little more than a nucleus, with almost all cytoplasm located in thickened branching processes. Extremely fine secondary branches are also present. This is the appearance of ramified microglia in tissue that is neither white matter nor gray matter (Case 17, CD45, ×770).

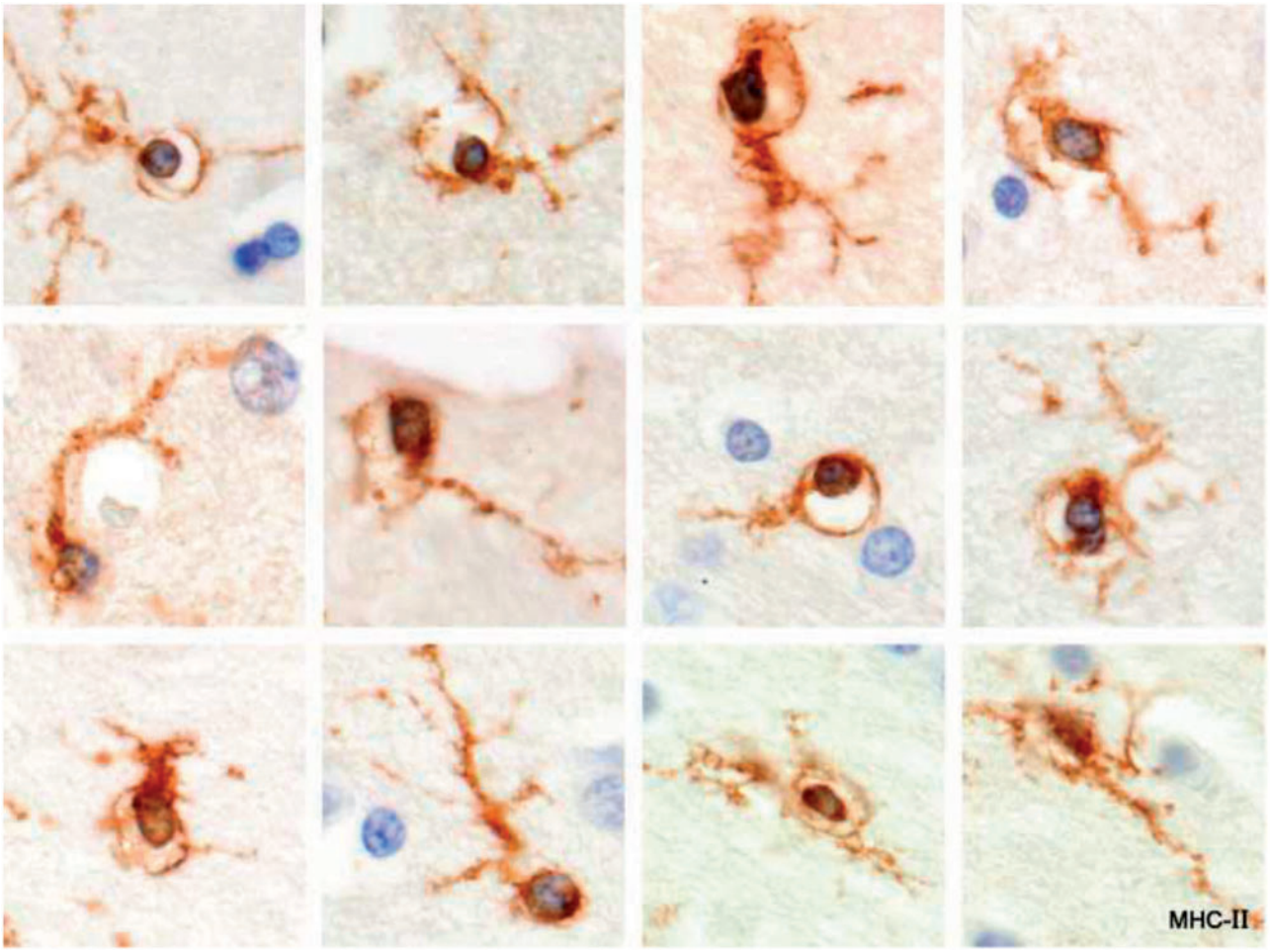

FIGURE 4. Myeloid cells, some ramified, corralling unidentified cells located in white matter bordering an expanding basal ganglia plaque. The enclosed cells may be degenerate. There is no evidence of phagocytosis (no phagosomes). (Case 4, MHC-class II, ×560).

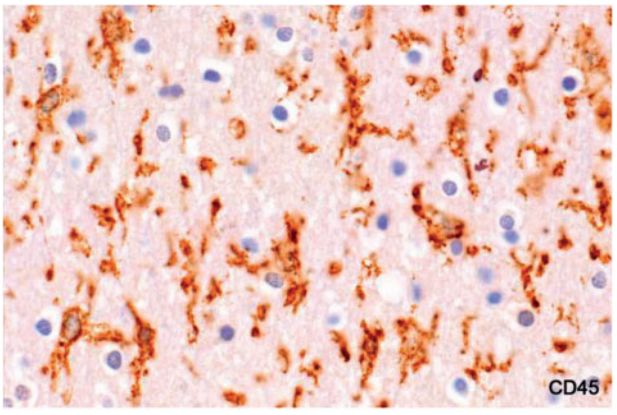

FIGURE 5. Early activated microglia. Numerous irregularly shaped microglia with thickened processes are present amongst normal-appearing oligodendrocytes in intact white matter remote from a plaque (Case 2, CD45, ×440).

monocytes located in capillaries in normal gray matter structures including the cerebral cortex was a common finding in most of the acute MS cases. The nodules described here, however, were observed only in Case 16, a patient with chronic progressive disease, ongoing recruitment of monocytes, and lesions with tight knots of unidentified MHC-class II-positive cells located in the glial wall of plaques. This is the second report of upregulation of MHC-class II antigen expression in lesion capillaries in MS (42–44).

Monocytes were present in large numbers, in and around all early lesions examined in 11 acute MS cases, in Case 7 and in Cases 16–18 (Fig. 10). Morphologically, these cells appeared to be typical CD45-positive monocytes; they were MHC-II-positive, MRP14-positive, and stained negatively for all lymphocyte markers tested. Monocytes were not detected in any of a large number of lesions examined in Case 19, a chronic case of 14 years duration with almost no lesions with lipid macrophages present in edge zones. Monocytes were lo-

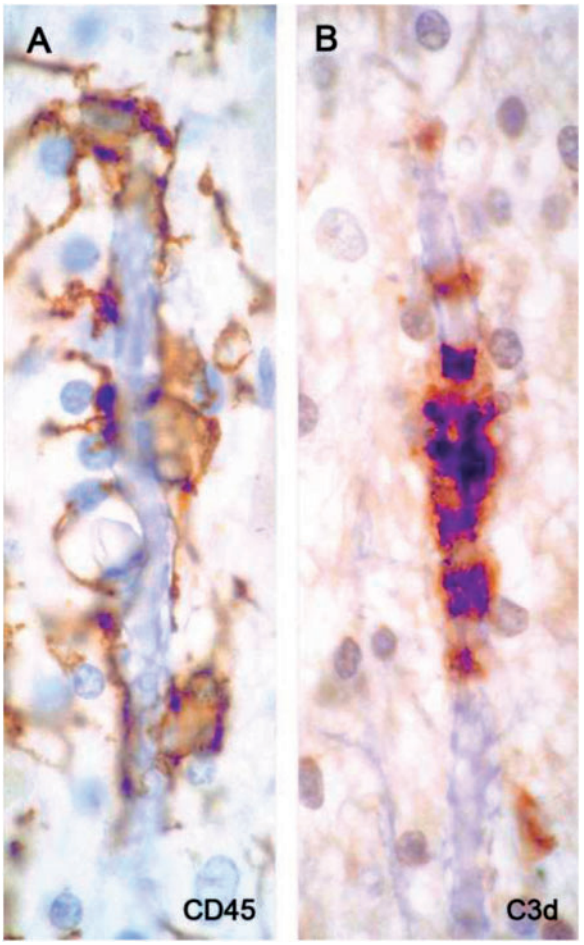

FIGURE 6. Microglial nodules centered on complement-positive myelin. **(A)** A nerve fiber engaged by a cluster of CD45+ elongated microglia. There is no evidence of phagocytosis. **(B)** A separate nodule stains positively for activated complement. Microglia show no complement-positive phagosomes (Case 16, **A**, CD45; **B**, C3d; **A, B** ×400). Reproduced from (39).

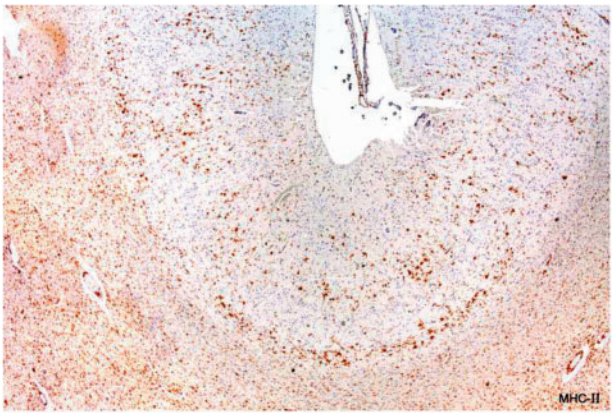

FIGURE 7. Microglia-monocyte-capillary nodules. The cerebral cortex is dotted with large numbers of MHC class II antigen-positive nodules. Higher power views are shown in Figures 8 and 9 (Case 16, MHC-class II, ×26).

cated in perivascular spaces, in the walls of small blood vessels, in the lumen of blood vessels, in actively demyelinating lesions including small perivascular lesions (Figs. 11–14 and Supplementary Data Figs. S5–S7), and as cuffs around vessels in the subarachnoid space, cerebral sulci and on the surface of the brain (Supplementary Data Fig. S8).

Mononuclear leukocytes underdoing mitosis were present in perivascular spaces in several cases where there were numerous PCNA-positive and Ki67-positive early macrophages nearby. The presence of monocytes in developing lesions including prephagocytic perivascular lesions was closely associated with the appearance in the tissue of myelin phagocytes as no lesion in any of the acute cases showed evidence of commencing myelin phagocytosis where they were absent. As with microglia, we were unable to identify an intermediate stage in their transition to phagocytes.

Perivascular cuffs composed entirely or almost entirely of monocytes were common. Lymphocytes were present in small numbers in perivascular spaces and in the parenchyma in all acute cases. Large cuffs predominantly of lymphocytes were also present but these were relatively rare. Importantly, with one exception, no polymorphonuclear leukocytes were seen in lesions of any age in any case. The single exception was the atypical Case 7 in which they were relatively numerous. No meningeal B-cell follicles containing plasma cells were identified in this series of acute cases (45). Plasma cells were absent in prephagocytic tissue and in all 6 actively demyelinating lesions examined in the 2 most acute cases in the series (Cases 1 and 2).

IgG-positive MRP14-positive MHC-class II-positive monocytes squeezed into elongated sausage-shaped forms in multiple capillaries in normal gray matter (pontine nuclei, cerebral cortex, granular layer of the cortex of the cerebellum) (capillary monocyte plugs) was a common finding in Cases 2, 4, 7, 9, 12, and 17 (Figs. 15 and 16).

Secondary Progressive MS

In 2 secondary progressive MS cases (Cases 16 and 17) and in Case 18, there was evidence of continuing recruitment of monocytes into and around plaques where active myelin breakdown was no longer evident (i.e., lipid macrophages present but no LFB-positive macrophages). The monocytes were located amongst lipid macrophages in demyelinated tissue, in the glial wall and in lesser numbers in the surrounding parenchyma (Fig. 17). The 3 cases were also unusual in having microglial nodules in bordering white matter; some of these contained not only early activated microglia but also monocytes.

Phagocytic Macrophages

Early macrophages, (cells referred to in routine diagnostic neuropathology as motile, ameboid, or pleomorphic microgliocytes or phagocytes), were located in areas of commencing myelin breakdown and had a histochemical profile typical of tissue histiocytes (Table 3). They also stained positively for IgG (Supplementary Data Figs. S9 and S10).

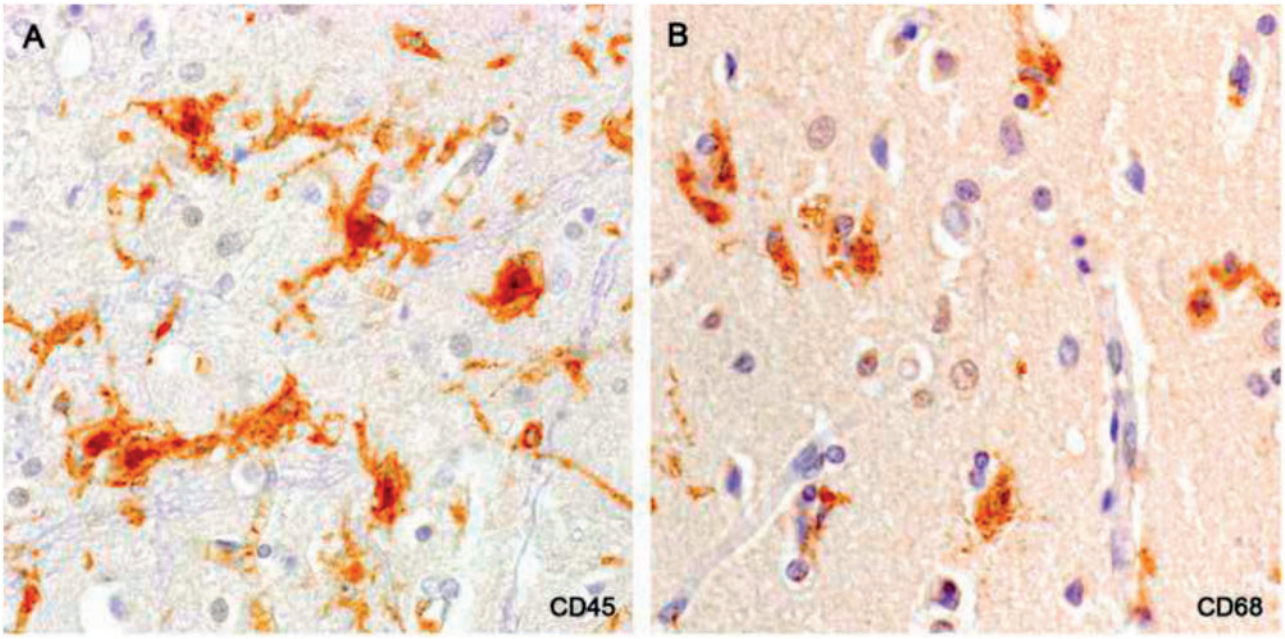

FIGURE 8. Microglia-monocyte-capillary nodules. Comparison with Figure 9 shows that MHC class II-positive capillaries are unstained in nodules stained for CD45 and CD68 (Case 16, **A**: CD45; **B**: CD68; **A**, **B,** ×560).

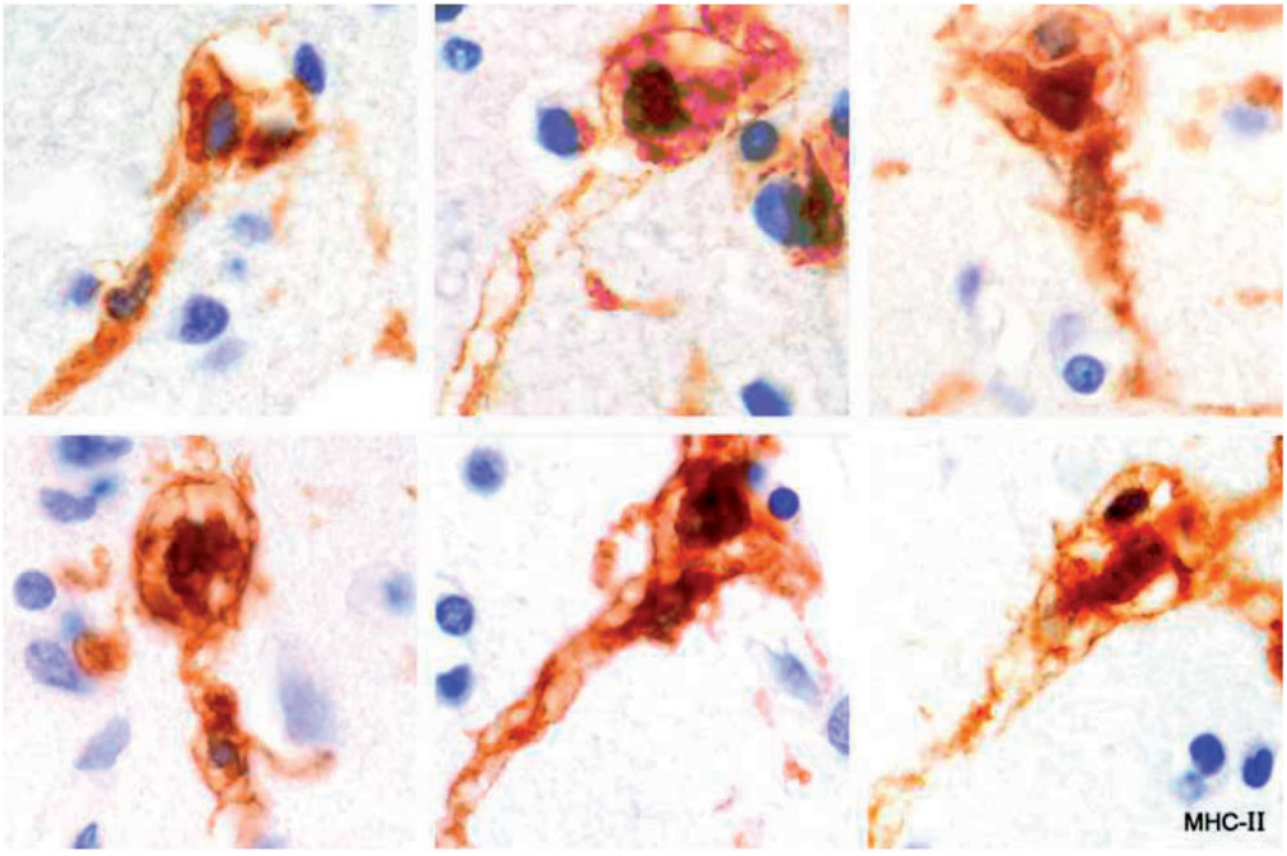

FIGURE 9. Microglia-monocyte-capillary nodules. Thin processes of ramified microglia enclose MHC-class II-positive cells associated with filmy, net-like MHC-class II-positive capillaries. Capillaries elsewhere in the cortex were MHC-class II-negative (Case 16, MHC-class II, ×560).

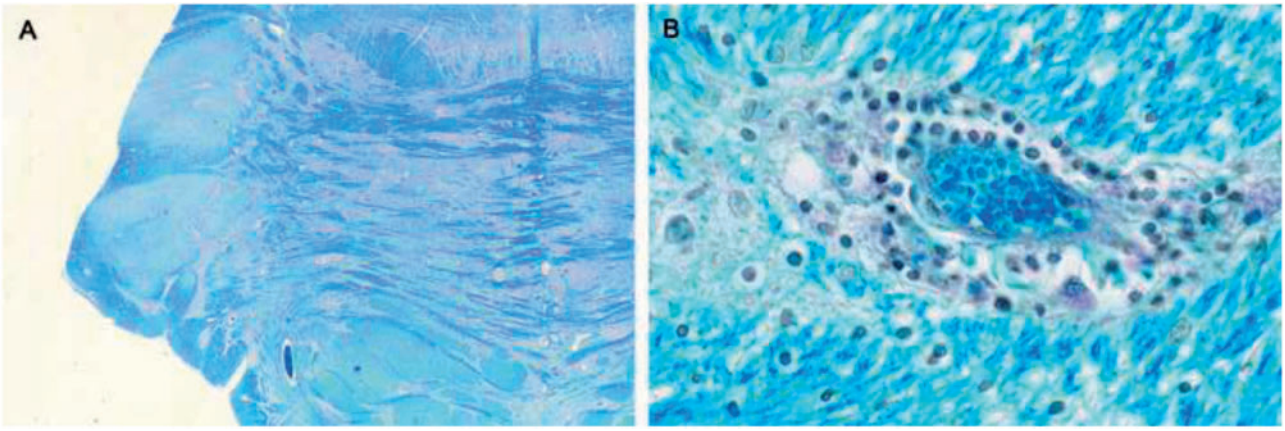

FIGURE 10. Perivascular monocytes. **(A)** Two large, circumscribed areas of pale but intact myelin are seen on the left of the figure. **(B)** Perivascular space mononuclear leukocytes are present around a vessel in one of the lesions. Higher magnifications showed few if any lymphocytes in the cuff (Case 12, **A, B**, Luxol fast blue; **A**, ×5, **B**, ×260).

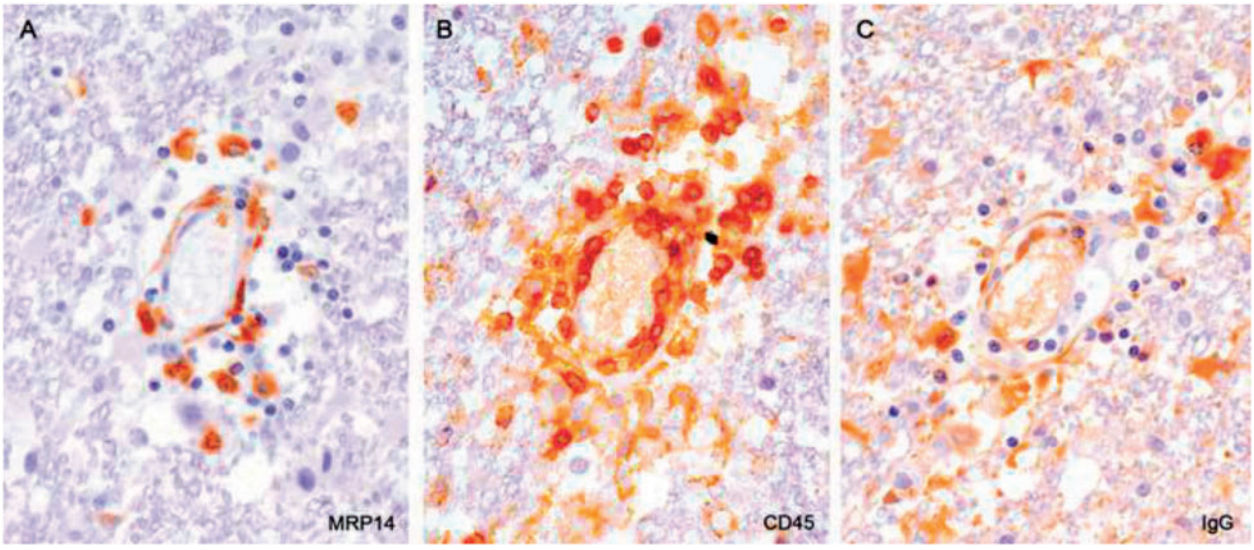

FIGURE 11. Monocyte infiltration. Skip serial sections of a blood vessel in intact tissue bordering a (postphagocytic) plaque. Apart from some vacuolation, myelin is intact. Monocytes are entering the parenchyma from the perivascular space where lymphocytes, if present, are inconspicuous. Extravascular IgG, chiefly in astrocytes, signifies an open blood brain barrier (Case 3, **A**, MRP14; **B,** CD45; **C**, IgG; **A–C**, ×230).

Contents ranged from no discernible phagosomes (Fig. 18) to large cells containing particles of myelin (Fig. 19). Myelin sheaths contacted by infiltrating phagocytes or in their immediate vicinity appeared spongy due in part to the presence of intramyelinic edema; they reacted positively for activated complement (C9neo) (Fig. 20); and they stained positively with biotinylated normal and MS CSF IgG. The latter was especially apparent near expanding lesions in Cases 3 and 10.

In partially demyelinated tissue close to the edge of the lesion illustrated in Figure 19 (Case 20), myelinated fibers contacted by myelin phagocytes showed sheaths of reduced thickness, focal lysis of superficial lamellae and detached fragments of compact myelin (Supplementary Data Fig. S12). Vesiculated myelin, an important change noted in some EM studies of macrophage-myelin engagement in MS, was not observed in this case. Counts of oligodendrocytes identified by EM in tissue bordering the edge of the same lesion (10) showed some reduction in oligodendrocyte numbers in areas where macrophages and lymphocytes were observed in contact with degenerate oligodendrocytes and unidentified cells (Fig. 21 and Supplementary Data Fig. S11).

None of the large number of lesions examined by EM in the longstanding case of MS (Case 19) showed evidence of recent myelin breakdown or the presence of monocytes. Plaque margins showed occasional small phagocytes with lipid

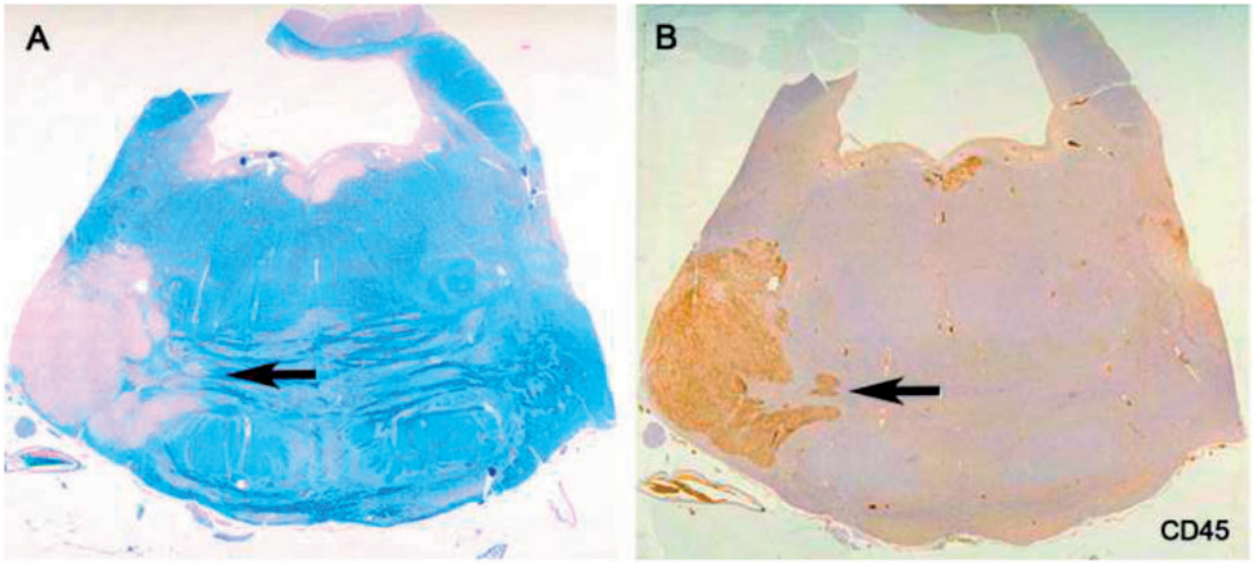

FIGURE 12. Exceptionally early lesions in exceptionally early multiple sclerosis. The lesion at the far right showed, at high magnification, pyknotic oligodendrocytes and pale but intact myelin. The lesion near the ventricle showed vacuolated but otherwise intact myelin. The large lesion on the left showed commencing loss of myelin in the presence of numerous early phagocytes and monocytes. The area indicated by the arrow is shown at higher magnification in Figure 13. Unidentified large IgG + mononuclear cells were present throughout the section (Case 2, **A**, Luxol fast blue-PAS; **B**, CD45; **A, B**, ×2.5).

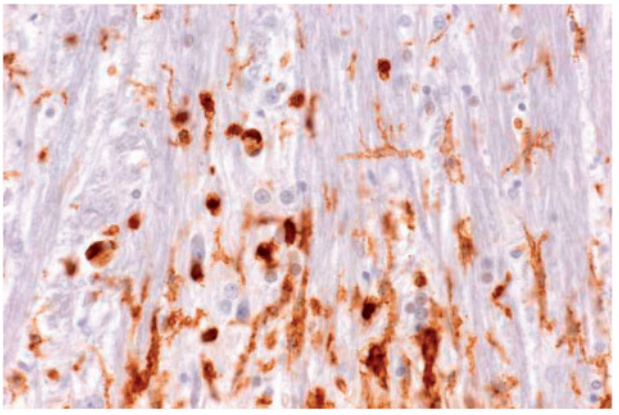

FIGURE 13. Edge of an expanding plaque. Microglia in the intact margin are of the nonactivated small, ramified phenotype. Rounded cells in the vicinity are monocytes. Whether the elongated irregularly shaped cells are early activated microglia or early macrophages of monocyte origin is unclear (Case 2, CD45, ×500).

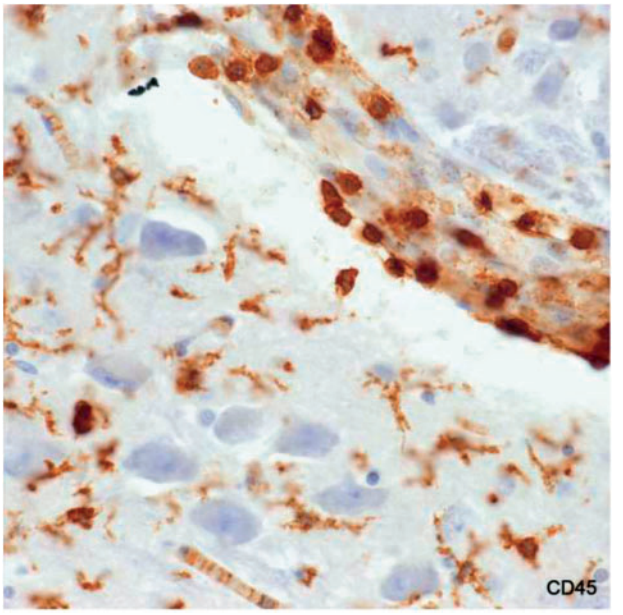

FIGURE 14. Infiltrating monocytes. A small blood vessel at the edge of the large developing lesion shown in Figure 12 is cuffed with monocytes. Microglial cells in the surrounding tissue are mostly ramified or early activated in type (Case 2, CD45).

vacuoles and secondary lysosomes composed of membrane bound stacks of distinctive filamentous material but no recognizable particles of myelin. A few such microglia/macrophages were seen attached via clathrin-coated pits to loosened myelin lamellae on the surface of otherwise normal appearing myelin sheaths (Fig. 22 and Supplementary Data Fig. S13).

An important difference in the 2 cases relates to the number of plasma cells present in lesions. In the actively demyelinating lesion in Case 20, no plasma cells were observed in the albeit small amount of tissue examined. In Case 19, where there was little evidence of recent demyelinating activity, counts of perivascular and parenchymal plasma cells determined in semithin epoxy sections showed plasma cells to be numerous in plaque tissue and in surrounding intact tissue. Actual numbers were 1772 plasma cells per cubic mm in

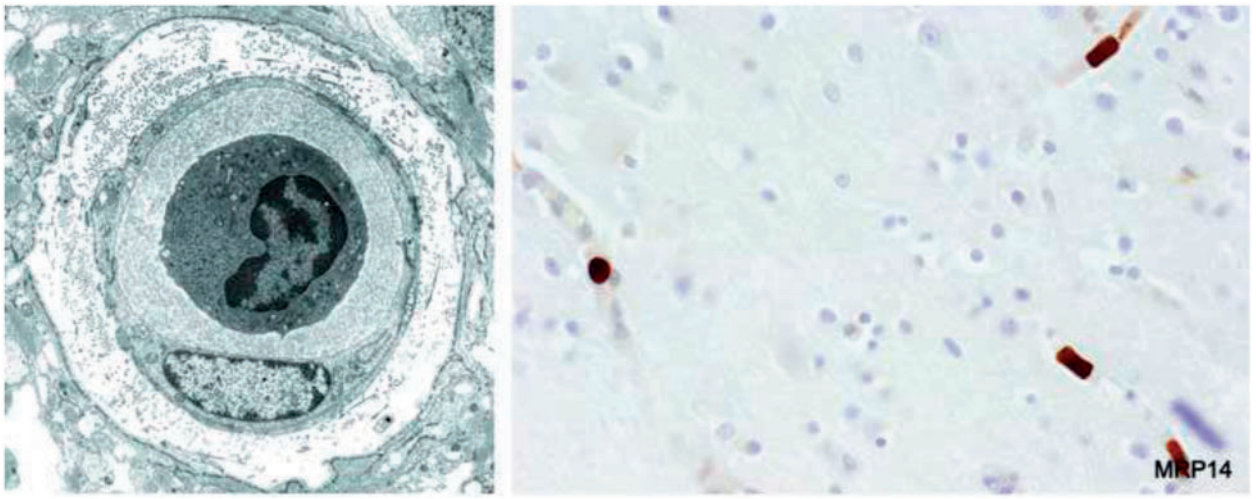

FIGURE 15. Monocytes. (Left) An intracapillary monocyte in an old white matter plaque. Typical of white matter capillaries there is a prominent perivascular space. (Right) Four squeezed monocytes (plugs) in normal gray matter capillaries in intact tissue remote from a plaque. (Left: Case 19, electron micrograph, original magnification: ×2800. Right: Case 7, MRP14, ×260).

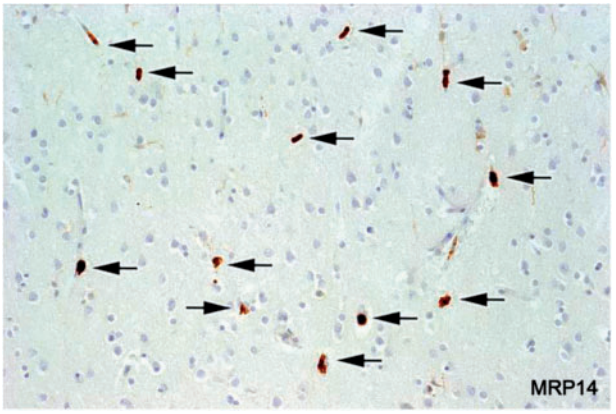

FIGURE 16. Monocyte plugs in normal gray matter. Those in capillaries appear squeezed (Case 7, MRP14, ×60).

demyelinated tissue and 389 per cubic mm in the surrounding white matter. Plasma cell counts in a control case of motor neuron disease showed no plasma cells (46, 47).

Lipid Macrophages

Lipid macrophages comprised a population of large IgG-positive cells (Fig. 23). These cells were swollen with Oil Red O-positive metabolized myelin and were located behind a phalanx of active phagocytes at plaque margins. In the case of nonenlarging plaques, (that is no LFB-positive macrophages), they were located in demyelinated tissue towards the plaque rim. Large fragments of MBP-positive, MAG-positive myelin were attached to the surfaces of some lipid macrophages. This population of cells showed clustering of surface IgG (Figs. 24 and 25) (48). These cells correspond to the lipid macrophages

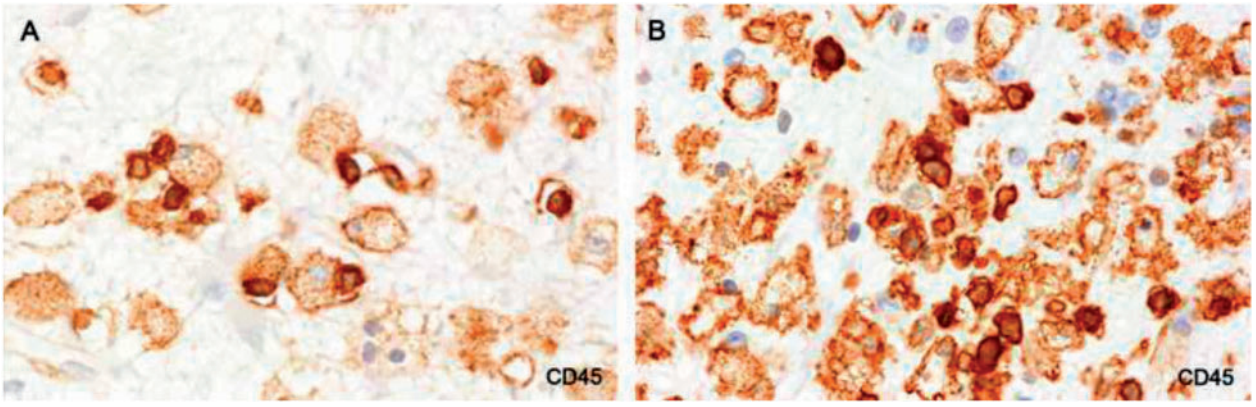

FIGURE 17. Secondary progressive multiple sclerosis. **(A, B)** Demyelinated tissue in 2 chronically inflamed nonexpanding lesions (no Luxol fast blue-positive macrophages) shows a mixture of lipid macrophages and recently recruited monocytes. Monocytes appear not to be distributed randomly with respect to the lipid macrophages, a disproportionate number locating close to a macrophage (**A**, Case 16, CD45. **B**, Case 17, CD45. **A, B**, ×240).

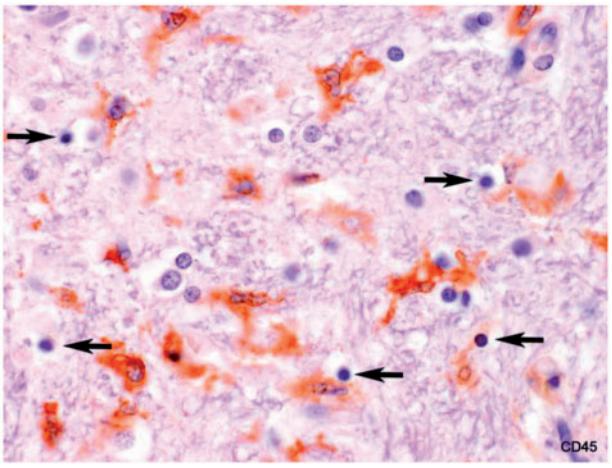

FIGURE 18. Commencing macrophage activity in pre-phagocytic multiple sclerosis tissue. Myelin appears to be intact, macrophages lack phagosomes, and there are some oligodendrocytes present with pyknotic nuclei (arrows) (Case 1, CD45, ×580).

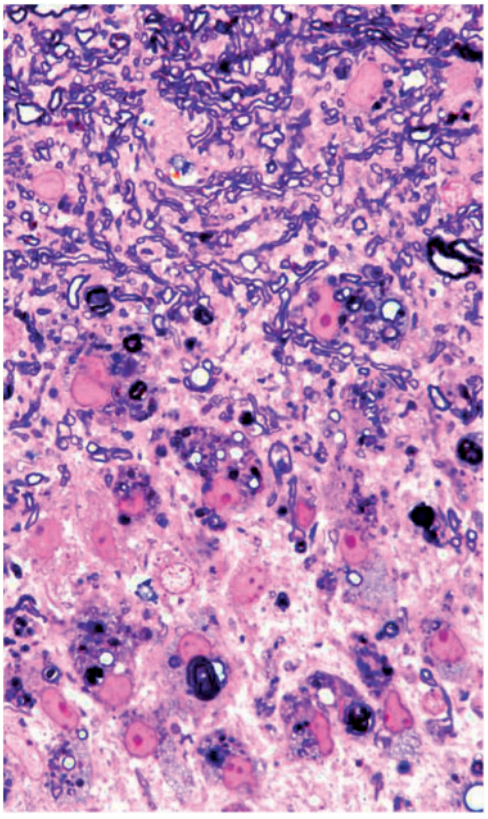

FIGURE 19. Active demyelination at the edge of an expanding lesion. Cell counts determined electron microscopically showed in normally myelinated bordering tissue (upper third of the figure) lysis of oligodendrocytes. The middle third of the figure shows myelin loss occurring in the presence of myelin phagocytes. The lower third of the figure is a trailing area of lipid macrophages with granular myelin debris (Case 20, semithin epoxy section, toluidine blue and safranin, ×1200).

designated M2 anti-inflammatory macrophages that have been described in postphagocytic plaques (50, 51).

Small Lipid Macrophages

Small spindle-shaped cells containing small amounts of lipid identified in frozen sections were observed in some of newly forming gray matter plaques. These cells correspond to the small lipid containing cells observed by Dawson in normally myelinated tissue bordering early lesions.

Macrophages and Oligodendrocytes

Microglia/macrophages phagocytosing apoptotic oligodendrocytes were observed in Case 9. In Case 20, macrophages were seen contacting degenerate oligodendrocytes (Supplementary Data Figs. S11 and S14).

Macrophages and Astrocytes

Phagocytosis of large stellate AQP-positive astrocytes in recently demyelinated tissue was observed in several of the cases of clinically early MS, as reported previously (38). This activity was selective in the sense that AQP4-negative gemistocytic astrocytes in the immediate vicinity were spared.

Residual Microglia

Microglia/macrophages with distinctive cytoplasmic inclusions consisting of membrane bound stacks of curved linear profiles were common in autopsy tissue examined by EM in Case 19. The same inclusion-bearing cells have been noted in MS by many authors and, according to some, in unrelated diseases. Usually located in partially myelinated tissue these cells are also seen crossing into perivascular spaces where they are phagocytosed by resident perivascular space macrophages. That the inclusions are metabolized myelin is unproven and it may be that that these cells are not myelin phagocytes but normally functioning microglia (52). This study provided little reliable immunohistochemical data related to these cells, particularly regarding CD45 and MHC-II antigen immunoreactivity.

Perivascular Space Macrophages

Macrophages located in this mesodermal compartment have unusually large primary lysosomes, and, in contrast to resident microglia, are actively phagocytic and express IgG Fc gamma receptors (53). In MS Case 19, they were seen phagocytosing debris-laden cells that had crossed the glial limiting membrane and entered the space.

Strangely Shaped IgG-Positive Mononuclear Leukocytes

The single instance where these were seen was in a brain stem section with 3 very early newly forming lesions in Case 2 (Figs. 12 and 26). They were present in perivascular spaces, in capillaries (plugs), and in the parenchyma near and at a dis-

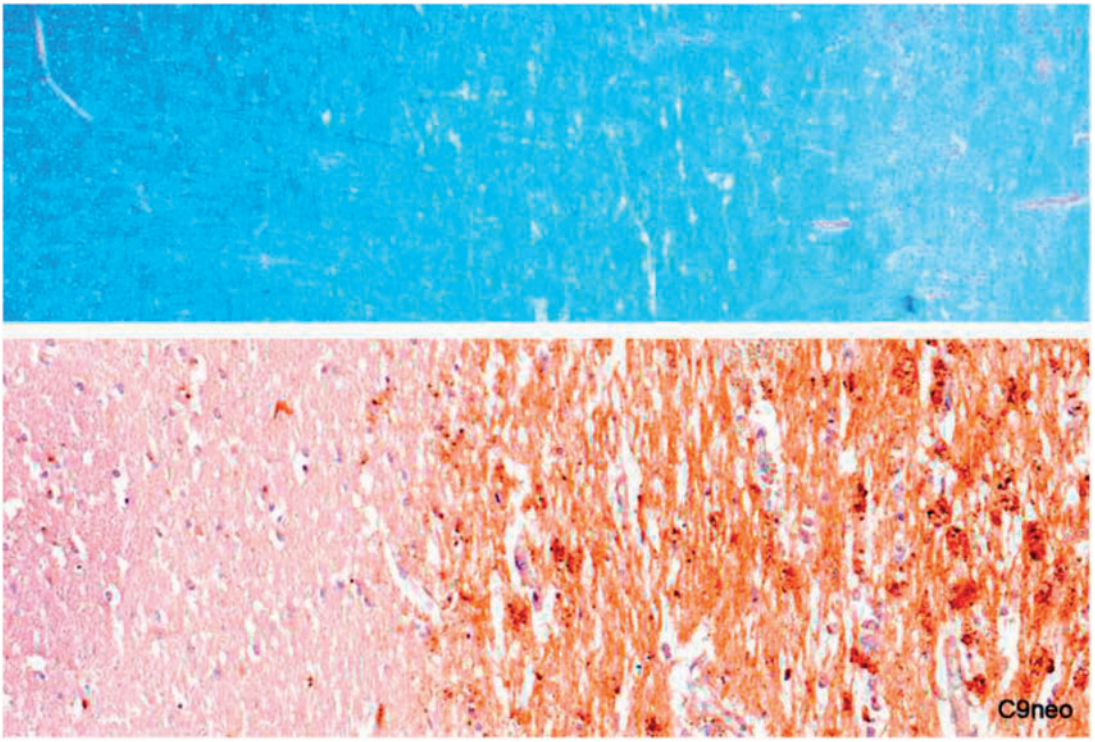

FIGURE 20. Myelin sheaths and activated complement. (Top Image) There is a broad zone (middle third of the picture) of vacuolated but otherwise intact myelin bordering the edge of an expanding plaque. Higher magnifications showed widespread intramyelinic edema and oligodendrocytes with pyknotic nuclei. (Lower Image) A similar pre-phagocytic lesion with oligodendrocyte nuclear pyknosis and loss shows myelin sheaths that stain positively for activated complement (Top, Case 2, Luxol fast blue, ×22. Lower, Case 3, Membrane Attack Complex, C9neo, ×80).

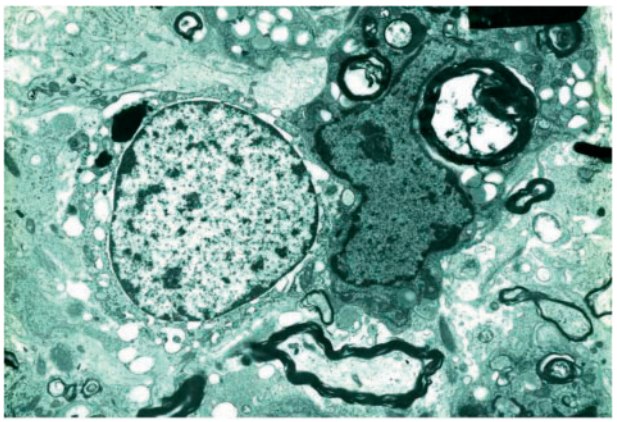

FIGURE 21. Prephagocytic microglia/macrophage activity. A degenerate glial cell with swollen cytoplasm and disrupted plasma membranes, possibly an oligodendrocyte, is contacted by a macrophage. Other degenerate glial cells in the same area were observed in contact with typical small lymphocytes. The figure is from tissue bordering the lesion illustrated in Figure 19 (Case 20, electron micrograph, original magnification: ×5000) Reproduced with permission from (24,).

tance from the 3 developing plaques. The cytoplasm stained evenly for IgG without edge accentuation. Except for those located in capillaries, the unusual shape was maintained whether they were located in open perivascular spaces or in compact tissue, Clinical and other details of the case are described in Supplementary Data Case 2.

Small Elongated MRP14-Positive Cells

Small elongated cells were detected in intact tissue bordering cerebral hemisphere lesions in a patient with a severe neuromyelitis optica (NMO) spectrum variant characterized by infiltration of affected tissues by both monocytes and neutrophils (Figs. 27–29; Supplementary Data Fig. S19, and Supplementary Data Case 7). The true size and shape of the cells could not be determined as the cells were identified only in sections stained for MRP14. In such sections, the only parts of the cell that were visible were those parts staining positively for MRP14.

CD209-Positive Dendritic Cells

These cells were common in the walls of small venules and in cuffs of inflammatory cells in newly forming MS lesions.

Microglia Undergoing Mitosis

Mitoses in microglia were observed in otherwise unaffected tissue in Case 7, a patient with a fulminant spinal and cerebral variant of NMO (Supplementary Data Fig. S18).

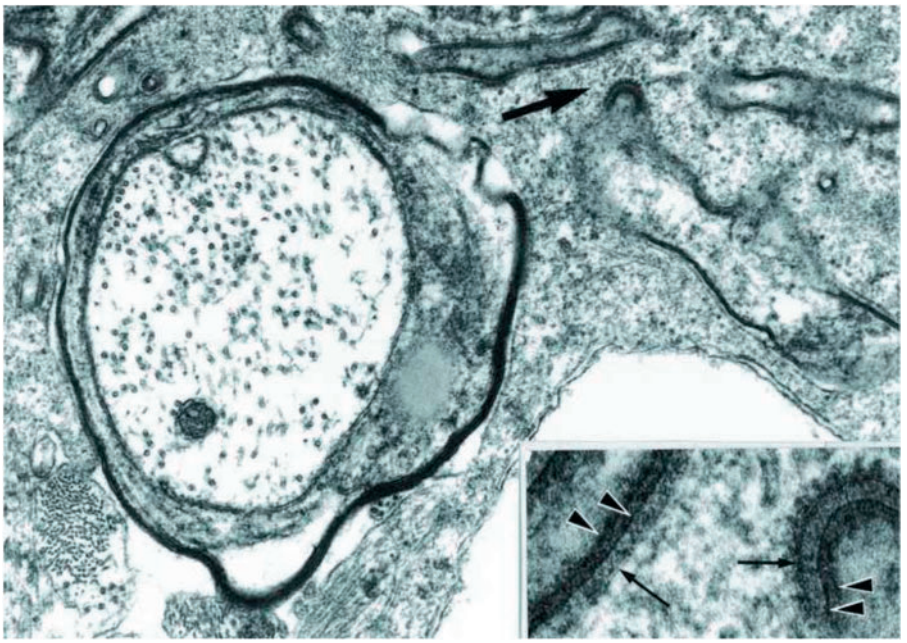

FIGURE 22. Micropinocytosis vermiformis. Loosened myelin lamellae are attached to clathrin-coated pits on the surface of a microglia/macrophage. The inset of the area indicated by the arrow shows myelin lamellae (arrow points) separated by a constant gap from the plasma membrane (small arrows) of the phagocyte (Case 19, electron micrographs, ×40 000, inset, ×170 000). Reproduced with permission from (9).

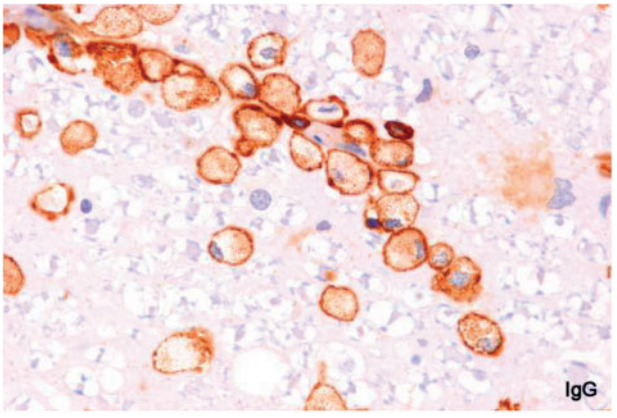

FIGURE 23. IgG-positive lipid macrophages in demyelinated tissue in a late postphagocytic plaque. Of interest is the fact that there is almost no IgG detectable in the tissue except for that present on the surface of the macrophages. Two monocytes and what may be an astrocyte also stain positively for IgG (Case 16, IgG).

Other CD45-Positive Myeloid Cells

These included macrophages contacting via clathrin-coated pits complement-positive mineral inclusions in the choroid plexus, and ramified microglia in contact with the glial limiting membrane around blood vessels and at the pial surface. No nodules of lymphocytes of the type seen in patients with paraneoplastic encephalomyelitis and antibodies against intracellular antigens were seen in any MS case (54).

Oligodendrocytes and Astrocytes

Sections of the medulla in Case 9 showed oligodendrocytes with fragmenting pyknotic nuclei located amongst normal looking nerve cells in intact myelinated gray matter close to the floor of the fourth ventricle and at a second site (Figs. 30–32 and Supplementary Data Fig. S3). The presence of apoptotic nuclear bodies located in tags of cytoplasm confirmed this to be classical apoptosis of oligodendrocytes. The location of degenerate oligodendrocytes amongst normal neurons in myelinated gray matter adds to existing evidence that oligodendrocyte loss in MS precedes and probably determines loss of myelin in MS. Nothing similar has been noted in studies of early postphagocytic plaques containing large numbers of oligodendrocytes. Other types of oligodendrocyte injury are illustrated in Supplementary Data Figures S11, S14–S16. Large astrocytes with pale cytoplasm and unusually pale nuclei were observed in intact myelinated tissue bordering some acutely expanding plaques (Supplementary Data Fig. S17).

Progressive Gliosis

Two patients with longstanding MS and no evidence of ongoing myelin breakdown (Cases 18 and 22) showed marked over expression of AQP4 in demyelinated tissue as well as in surrounding intact tissues. In 1 of the 2 cases, this was exaggerated to the point that affected plaques were indiscernible in sections stained for AQP4.

DISCUSSION

Using a staining procedure considered by some to be specific for microglia, del Rio Hortega and others reported

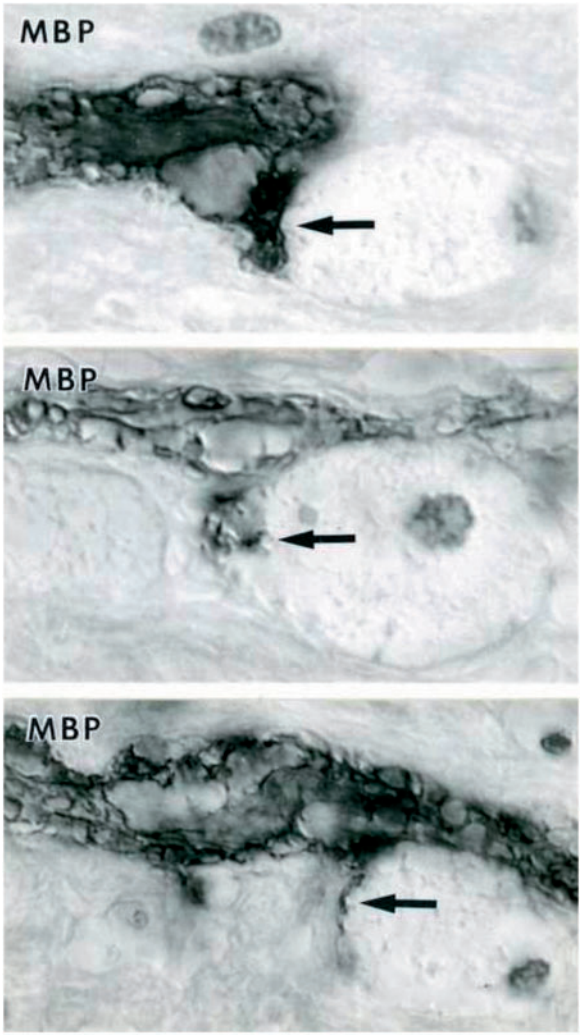

FIGURE 24. Arrested phagocytosis. Myelin basic protein (MBP)-positive myelin fragments partially detached from myelin sheaths are shown attached to the poles of lipid macrophages contacting sheaths (Case 16, MBP and Nuclear Fast Red counterstain, ×1300). Reproduced with permission from (49).

that phagocytes including lipid macrophages that are present in focal brain lesions of various sorts develop from resident small branching microglia. There is an equally longstanding alternative view, namely that brain macrophages develop chiefly from infiltrating monocytes and other leukocytes. This study shows that monocytes are a major source of phagocytes in MS lesions.

Microglia and Other Myeloid Cells

This study identified 20 myeloid cell subtypes or categories including 2 cell types not known previously to occur in demyelinating diseases. The following is a classification of CD45-positive myeloid cells in lesions of different histological age in patients with early and late MS: (i) Microglia - resident, reactive, mitotic, and residual. (ii) Microglial nodules - C3d-positive, monocyte-positive, and capillary-positive. (iii) Monocytes - capillary plugs, vessel walls, perivascular spaces, parenchyma, and nodules. (iv) Macrophages phagocytic - of myelin, oligodendrocytes, and astrocytes. (v) Macrophages nonphagocytic - lipid. (vi) Mesenchymal macrophages -perivascular spaces, meninges, and choroid plexus. (vii) Strange IgG-positive mononuclear leukocytes. (viii) Small elongated MRP14-positive cells in a patient with an NMO variant.

Macrophage-Mediated Demyelination

MS plaques, small perivascular lesions and subpial strips of demyelination, the 3 main forms of focal myelin loss in MS, develop in relation to perivascular infiltrates of inflammatory cells, initially chiefly monocytes together with lymphocytes and other inflammatory cells. Myelin loss is selective, leaving axons relatively intact. There is evidence that this selective loss is caused not by oxidative stress or some other nonspecific effect of aggregates of inflammatory cells but to a particular property of the inflammatory response in MS (55).

Although the event that initiates commencing loss of myelin is unknown, the proximate cause of myelin destruction involves the appearance in the tissue of what seems to be a homogeneous population of early IgG-positive Fcγ receptor-bearing macrophages in tissue that normally lacks such cells and in which there is no IgG (56, 57). Accompanying changes present from the beginning include signs of a disrupted blood-brain barrier, deposition of activated complement and other degenerative changes in oligodendrocytes and myelin, loss of astrocyte foot processes, and an increase in number, size and shape, and other evidence of activation of microglia. The population of active phagocytes changes in time into a population of nonphagocytic lipid macrophages that exit the plaque via mesodermal perivascular spaces.

Little is known about this population of early phagocytes except that the cells are motile, IgG-positive myeloid-derived macrophages that appear in developing lesions at the same time as recruited monocytes. It can be assumed that they are Fcγ receptor-positive (lipid macrophages are FcR-positive) (56, 57), but there are no genomic or other identifies known at this time.

Regarding the origin of early phagocytes, whether largely from monocytes or from both microglia and monocytes remains uncertain (58). Both cell types are present together in the parenchyma at the time early phagocytes begin appearing, with both disappearing as the proportion of phagocytes increases. How they combine, assuming that this is what happens, to produce the population of early macrophages is unknown. Although the rounded monocyte-type cells in perivascular spaces and in the parenchyma are likely monocytes from the circulation, it is difficult to be absolutely sure that all large active phagocytes originate from monocytes rather than transitioning rounded or ameboid cells of microglial origin.

Myelin Autoantibodies

The current view, that macrophages interact with myelin in MS under the direction of T lymphocytes, is based largely on the EAE model as reviewed by Hohlfeld et al (59, 60). That

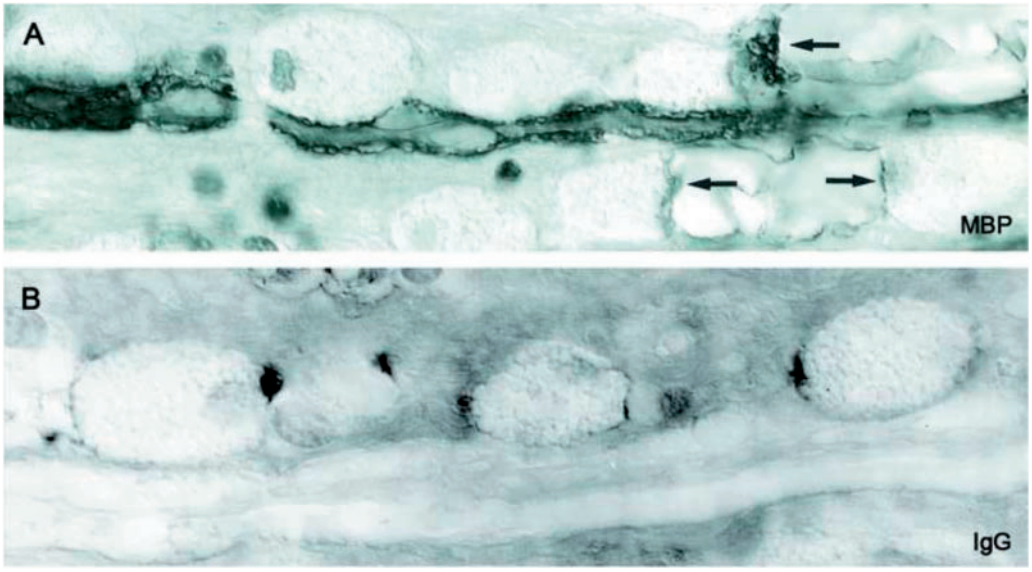

FIGURE 25. Arrested phagocytosis. **(A)** Lipid macrophages contacting a still largely intact myelinated nerve fiber have myelin fragments attached to the ends of the cells. **(B)** A section from the same region of the plaque stained for IgG shows surface IgG on aligned lipid microglia/macrophages in the form of polar caps (Case 16, **A**, MBP, ×700; **B**, IgG, ×890). Reproduced from (48).

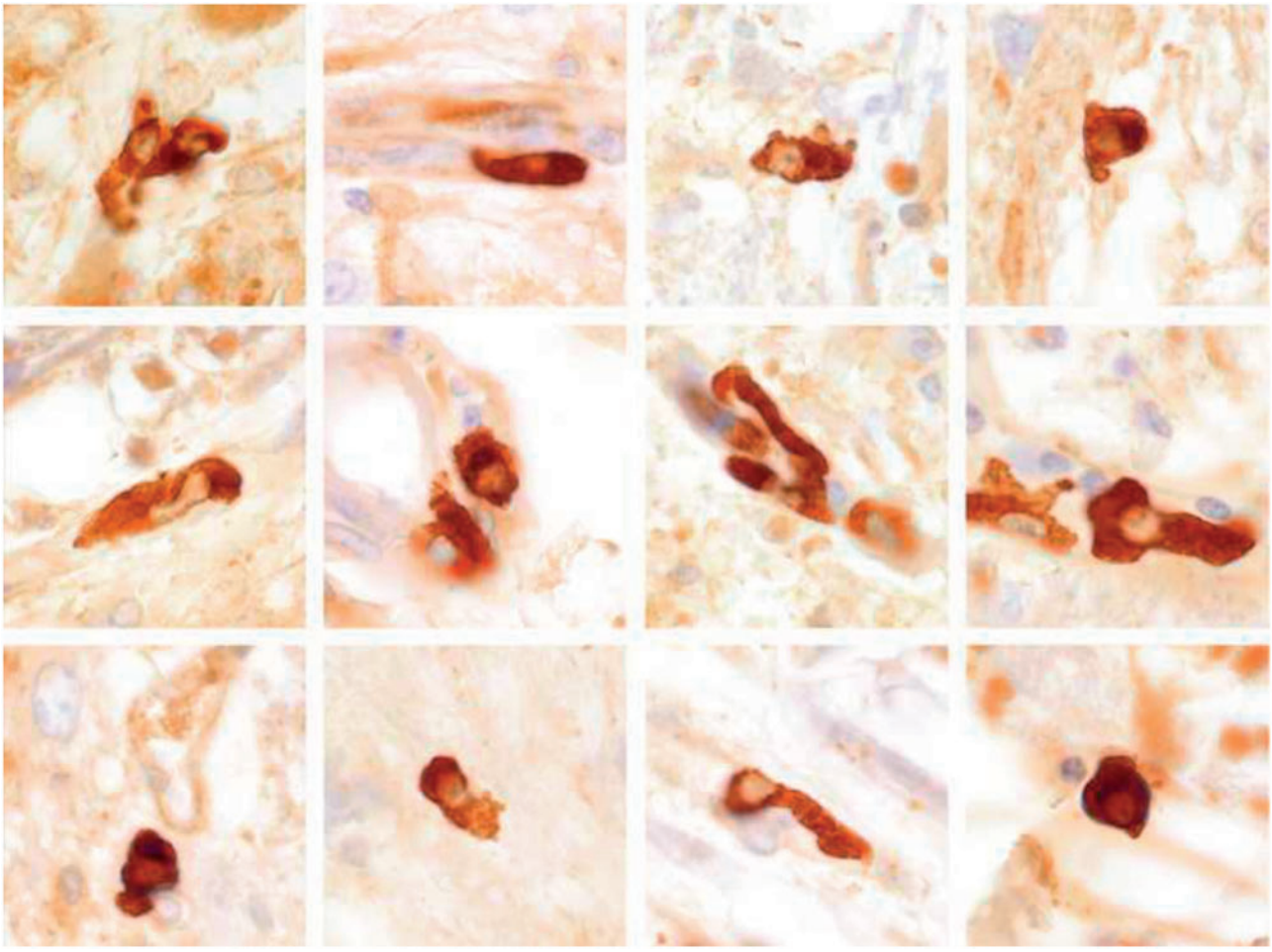

FIGURE 26. Unidentified IgG-positive mononuclear leukocytes. These were present in perivascular spaces, as "capillary plugs," and in the parenchyma, in and around newly forming lesions in the medulla in an exceptionally early case of multiple sclerosis (Case 2, IgG, ×720).

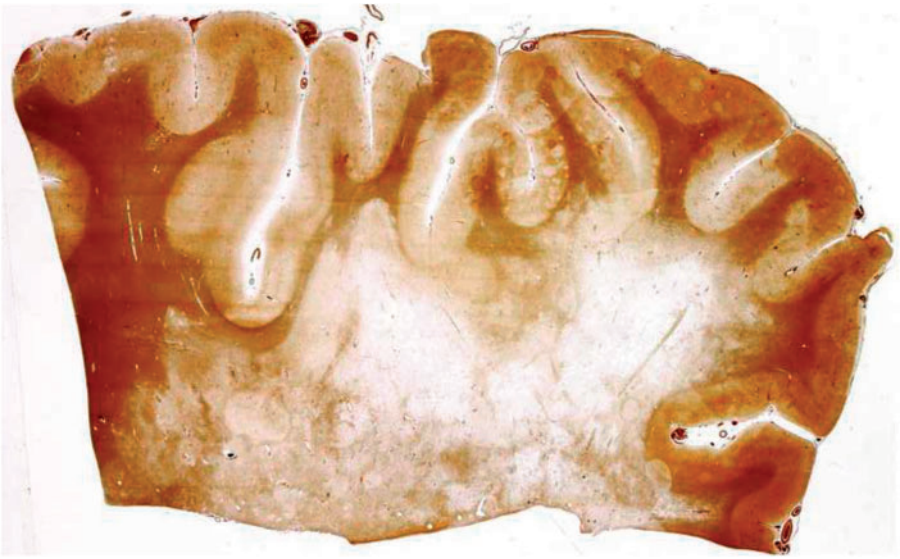

FIGURE 27. Confluent perivascular demyelinating and destructive hemisphere lesions in a patient with an neuromyelitis optica variant (Case 7, Glees silver impregnation, ×2.0).

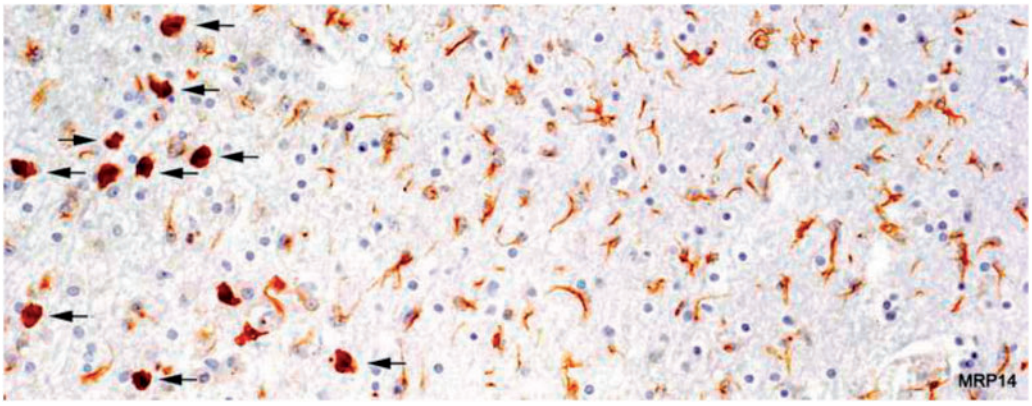

FIGURE 28. Unusually shaped small myeloid cells expressing the activation marker MRP14. Intact tissue bordering demyelinated tissue infiltrated by MRP14+ monocytes and granulocytes (arrows) in a patient with an neuromyelitis optica variant (Case 7, MRP14).

specific antibodies may play a role is suggested by the reduction in clinical exacerbations seen in patients receiving treatment with monoclonal antibody therapies targeting the B-cell antigen CD20 (61).

Perivascular spaces in old MS lesions contain lymphoid-like tissue consisting of plasma cells, reticular cells, macrophages, lymphocytes, and lymphocyte–macrophage contacts resembling a type of immune synapse (46, 47). The same structures, termed ectopic lymphoid-like B cell follicles, are present in the leptomeninges and cerebral sulci close to demyelinated tissue on the surface of the brain, especially in cases of secondary progressive MS (45, 62, 63) leading to the view that these plasma cell aggregates act as a source of pathogenic autoantibodies that contribute to the formation of new plaques, perivascular lesions and subpial strips of demyelination (64). Against this is the fact that there are no plasma cells in most newly forming lesions and it may be that the clinical improvement accompanying treatment with anti-CD20 monoclonal antibodies is due not to suppression of local antibody production by plasma cells but to some other mechanism.

The present findings are consistent with reports that myelin sheaths in some early MS lesions stain positively for activated complement (C9neo). The study also shows that in fixed tissue myelin sheaths in tissue bordering areas of active demyelination bind biotinylated normal IgG and MS CSF IgG.

IgG Fcγ Receptor-Dependent Mechanisms

If the IgG from lymphoid-like structures or other sources is a pathogenic autoantibody its mode of action (MOA) is likely to be antibody-dependent cellular phagocytosis (ADCP) (65). Other "classical pathway" MOAs, that is complement-dependent cyto-

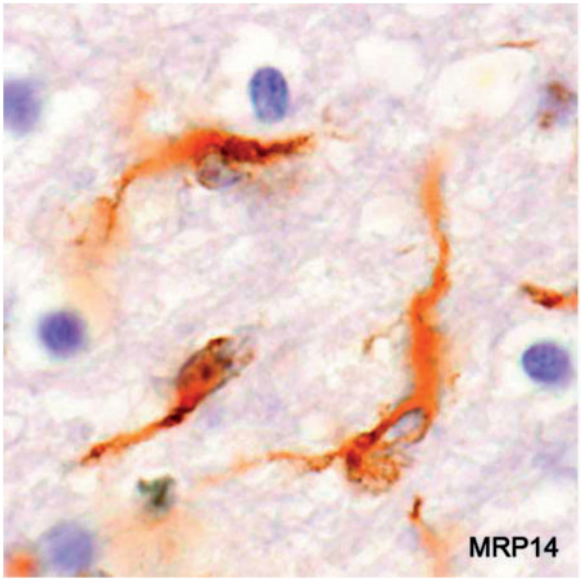

FIGURE 29. MRP14-positive atypical myeloid cells (Case 7, MRP14, ×1030).

toxicity, antibody-dependent cellular cytotoxicity, and programmed cell death (apoptosis) following binding of the antibody to the surface of the target cell (PCD), are other possibilities (66, 67). Activation of FcRs following Fc engagement by macrophages requires clustering of FcRs and the displacement of inhibitory receptors. Clustering of IgG on macrophages does occur in MS, which supports a role for ADCP in the disease.

Innate Immunity

On present evidence it is not known if the population of early macrophages involved in the destruction and removal of degenerate myelin, oligodendrocytes, astrocytes, and neurons in ischemic infarcts or traumatic brain injury differs from the population of phagocytes that effect destruction of myelin in MS. In experimental brain damage where the blood-brain barrier is disrupted, studies show that the great majority of phagocytes that invade for example, a small stab wound are monocytes that transform rapidly into early phagocytes as they emerge from blood vessels and later transform into typical nonphagocytic lipid macrophages (68, 69). In MS, selective interaction via innate immune mechanisms of macrophages with myelin sheaths and not with other cells and cell membrane in the same location, could be accounted for by changes in myelin determined by an oligodendrocyte lesion with the resulting uptake of degenerate myelin by phagocytes utilizing scavenger and complement receptors.

Bystander Demyelination

NMO is an inflammatory demyelinating disease affecting initially optic nerves and the spinal cord. During the course of the disease autoantibodies of different specificities sometimes develop. These include lupus antibodies, Sjogren's antibodies, AQP4 antibodies, and MOG antibodies and it is the characteristics of the autoantibody that determines the clinical and pathological features that distinguish each of the several NMO variants. Demyelination with axonal preservation in some NMO cases is associated with conspicuous oligodendrocyte apoptosis (70). It is possible that in MS the cause of demyelination and oli-

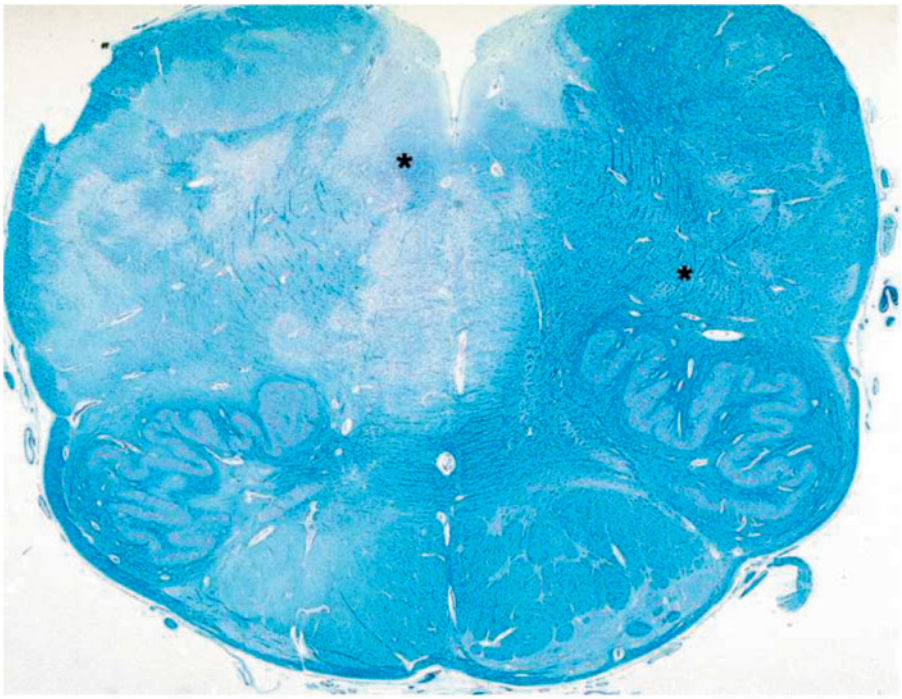

FIGURE 30. A medullary lesion, symptomatic for <24 hours, in a patient with relapsing and remitting multiple sclerosis. The asterisks indicate sites where apoptotic oligodendrocytes were detected in intact gray matter (Figs 31 and 32) (Case 9, Luxol fast blue, ×4.4).

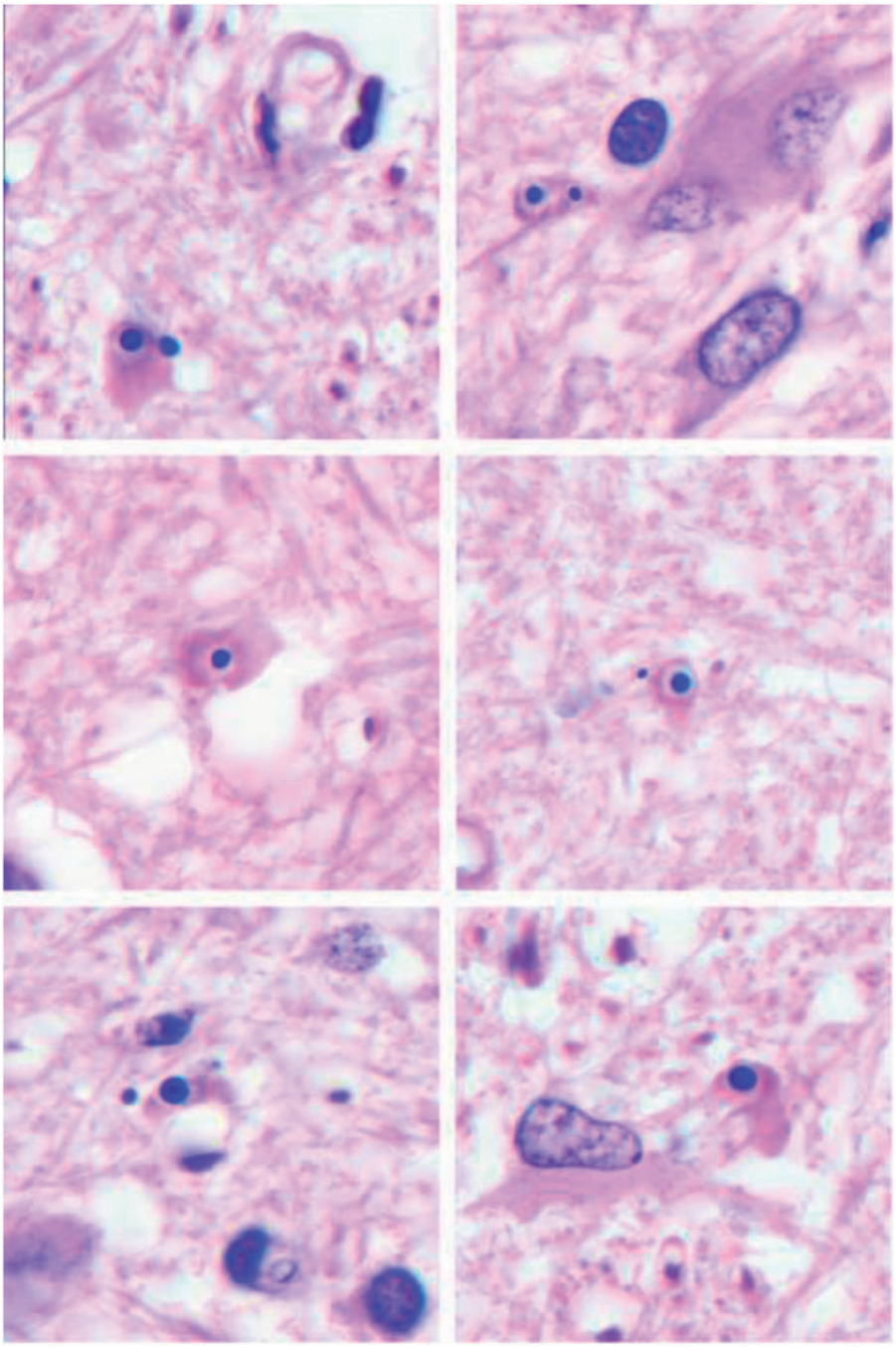

FIGURE 31. Apoptotic bodies in intact tissue bordering an expanding lesion (Fig. 30). Nuclear fragments in small tags of cytoplasm are located in tissue where oligodendrocytes are sparse or absent but other cells, that is astrocytes and neurons, appear normal (Case 9, hematoxylin and eosin, ×890).

godendrocyte apoptosis is not mediated by an adaptive immune mechanism targeting oligodendrocytes and myelin, but to some mechanism similar to that responsible for demyelination in NMO.

Secondary Progressive MS

The nodules composed of monocytes, MHC-II-positive capillaries and microglia observed in the cerebral cortex in Case 16 can be added to the list of gray matter lesions associated with progressive disease (71). Sobel et al in an electron immunocytochemical study of MS biopsy tissue noted that capillaries near white matter lesions in patients with clinically active disease but not in patients with inactive disease were immunoreactive for class II MHC antigens. MHC-II-positive capillaries were also noted in initial stages of lesion formation in EAE (42–44). These findings implicate this phenomenon of upregulation of the expres-

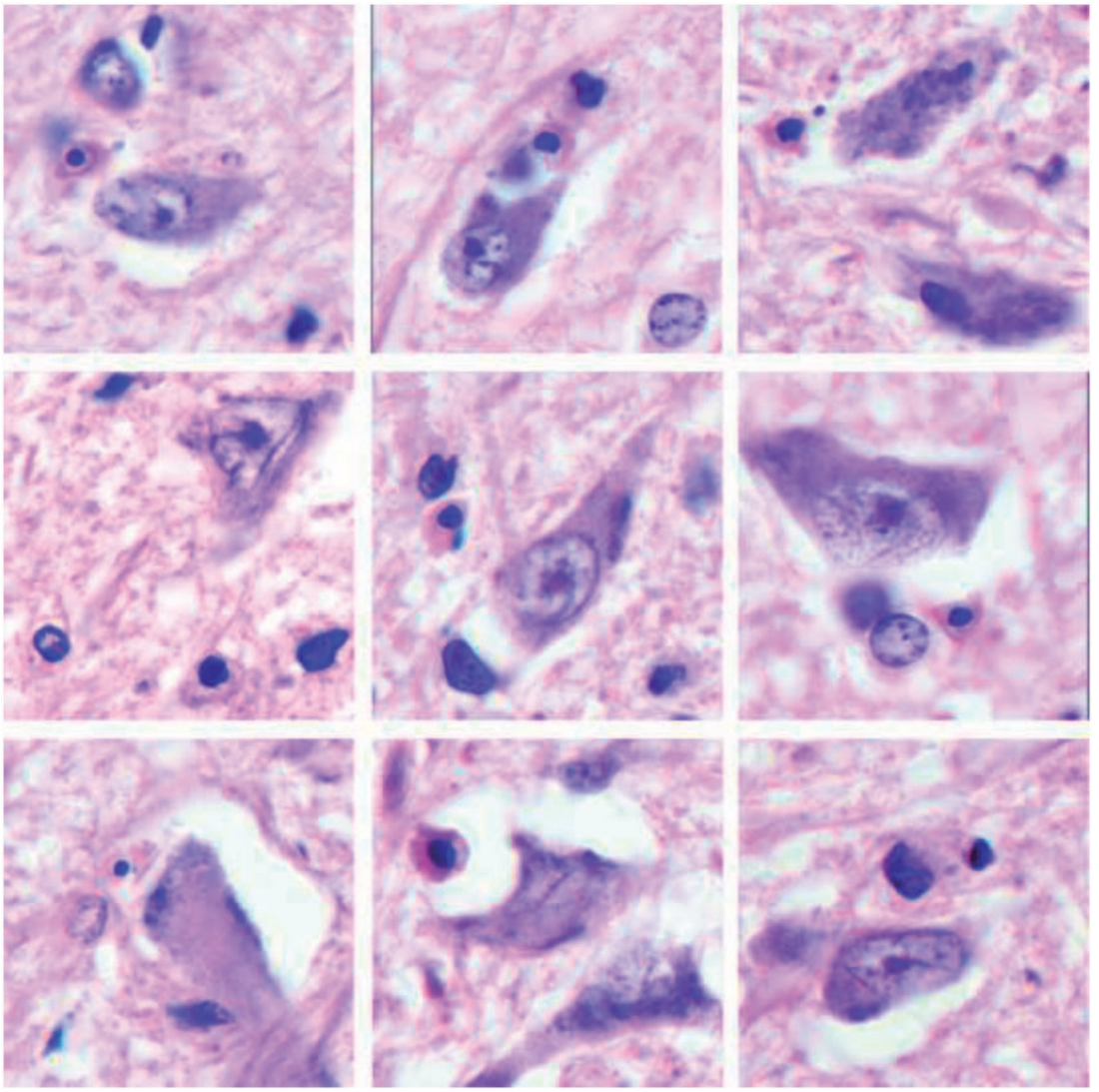

FIGURE 32. Apoptotic bodies in intact gray matter bordering an expanding plaque. Nerve cells in the same area are normal in appearance. The space around each nerve cell is a common post-mortems artifact (Case 9, hematoxylin and eosin, ×890).

sion by capillaries of MHC-II antigens in the pathogenesis of gray matter lesions in secondary progressive MS.

Monocyte Encephalopathy

Misshapen monocytes that is, plugs, located in capillaries in normal-appearing gray matter in most of the patients with early MS were present in numbers large enough to raise the possibility of compromised capillary circulation in affected tissues. Normal monocytes are larger than other leukocytes measuring between 16 and 22 μm in diameter (neutrophils are 9–15 μm in diameter). The diameter of a capillary lumen, on the other hand, is 3–10 μm "...barely wide enough for an erythrocyte to squeeze through" (72). The mismatch may be even greater in conditions where monocytes are atypical (73). Measurement of the diameter of capillaries and other small blood vessels in Case 19 using semithin toluidine blue-stained epoxy sections (46) showed that vessels measuring <23 μm in diameter were more than twice as numerous in cortical gray matter than in subcortical white matter, which could account for plugging affecting chiefly gray rather than white matter.

The apparent absence of any reactive tissue changes associated with the presence of large numbers of monocytes in cortical capillaries in early active MS suggests that any disturbance in function that might be associated with their presence is minor or transient. However, in Case 16, a patient with secondary progressive MS, microglial nodules centered on abnormal capillaries containing monocytes were present in numbers large enough to suggest the possibility of more serious cerebral cortical dysfunction.

The phenomenon of squeezed monocytes in gray matter capillaries present in numbers large enough to be of possible clinical significance is not restricted to MS. In Case 7, a patient with a severe progressive NMO variant (Supplementary Data Case 7) and who developed severe unexplained cognitive deficits terminally, monocyte capillary plugs in the cerebral cortex were excep-

tionally common. Whether other conditions with increased numbers of circulating activated monocytes, unexplained cortical dysfunction and normal MRI imaging show monocyte capillary plug formation (74) is yet to be determined.

Remodeling MS

In lesions sampled very early in their formation, (i.e., within hours or a few days), myelin loss is accompanied by a loss of oligodendrocytes and astrocytes. This suggests that the disease may not be a disease associated with a mechanism that specifically targets myelin.

In patients with longstanding disease, failure of repair of the oligodendrocyte lesion is common. Failure of repair of the astrocyte lesion is also not uncommon, the latter manifesting itself as a permanent opening of the blood-brain barrier and ongoing progressive gliosis.

In inflammatory demyelinating diseases in the peripheral nervous system, myelin destruction by macrophages occurs in the absence of microglia; in MS, 2 cell types seem to be involved in the process. It is uncertain regarding the degree, or the manner microglia contribute to the population of myelin phagocytes. The occurrence of microglia corralling monocytes in secondary progressive MS suggests differing roles for these 2 cell types in disease pathogenesis.

Summary

Myelin breakdown is initiated by a population of IgG-positive macrophages contacting largely intact myelin sheaths that stain positively for activated complement (C9neo). The appearance of this population occurs in the presence of a disrupted blood-brain barrier and is associated closely with commencing recruitment into the tissue of IgG-positive blood monocytes. Microglia and early activated microglia, in the absence of recruited monocytes, are nonphagocytic. How monocytes combine with nonphagocytic microglia to generate a population of phagocytes is unclear. The result, however, is the abrupt appearance in tissue that normally has no IgG and no Fc receptor-bearing cells of a large apparently homogeneous population of IgG-positive Fc receptor-bearing phagocytes that target myelin sheaths, apoptotic oligodendrocytes and astrocytes but leave nerve cells and axons relatively untouched. It is unclear how much of this macrophage activity is mediated by innate and adaptive immune mechanisms.

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