Relation between humoral pathological changes in multiple sclerosis and response to therapeutic plasma exchange

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Early, active multiple sclerosis lesions show four immunopathological patterns of demyelination. Although these patterns differ between patients, multiple active lesions from a given patient have an identical pattern, which suggests pathogenic heterogeneity. Therapeutic plasma exchange (TPE) has been successfully used to treat fulminant demyelinating attacks unresponsive to steroids. We postulated that patients with pattern II would be more likely to improve after TPE than those with other patterns since pattern II lesions are distinguished by prominent immunoglobulin deposition and complement activation. We retrospectively studied 19 patients treated with TPE for an attack of fulminant CNS inflammatory demyelinating disease. All patients with pattern II (n=10), but none with pattern I (n=3) or pattern III (n=6), achieved moderate to substantial functional neurological improvement after TPE ($p<0·0001$). Patients with multiple sclerosis with pattern II pathology are more likely to respond favourably to TPE than are patients with patterns I or III.

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Multiple sclerosis is an inflammatory demyelinating disease of the CNS, often associated with relapses. Although current treatments are uniformly applied, clinical, radiological, and pathological features are heterogeneous. Previous studies have described four immunopathological patterns of demyelination in early multiple sclerosis lesions characterised by inter-individual heterogeneity, but intra-individual homogeneity. Pattern I is characterised by T-cell/macrophage-associated demyelination; pattern II is characterised by antibody/complement-associated demyelination; pattern III is defined by a distal oligodendrogliopathy; and pattern IV is characterised by oligodendrocyte degeneration in periplaque white matter.[1] These observations could be clinically relevant since distinct patterns might need distinct treatments. Furthermore, since antibodies and complement contribute to multiple sclerosis pathology,[2] treatments to deplete them, such as therapeutic plasma exchange (TPE), could be effective.

About 45% of patients with fulminant attacks of multiple sclerosis and other idiopathic inflammatory demyelinating diseases (IIDD) of the CNS that are unresponsive to corticosteroids, improve after TPE.[3] Factors variably associated with favourable response include male sex, retained reflexes, and early treatment.[4] The response to TPE seems to be all or none, which suggests that a single biological factor might explain much of the variation. We postulated that immunopathological heterogeneity could contribute to differences in TPE response.

An inclusion criterion was TPE administered for a fulminant IIDD attack of the CNS resulting in major neurological deficit in one or more of the following: motor, cerebral, brainstem or cranial nerve, cerebellar, or sensory functions. Other criteria were sufficient clinical documentation to gauge response to therapy; tissue available from biopsy done before TPE to exclude alternative diagnoses, such as tumour, or from autopsy; pathology consistent with active inflammatory demyelination; and lesions immunopathologically classifiable into patterns I–IV, as previously described.[1] Exclusion criteria were: diagnoses of neuromyelitis optica, acute disseminated encephalomyelitis, or other non-multiple sclerosis disease on follow-up; and use of multiple concomitant immunosuppressive or immunomodulatory medication at time of TPE.

Insufficient tissue was available for immunopathological classification for three patients. 23 individuals met inclusion criteria, of whom four were excluded (two acute disseminated encephalomyelitis, one lymphoma, one multiple concomitant medication). 19 were included (16 biopsies, two both biopsy and autopsy, and one autopsy). Long-term clinical follow-up was obtained for 16 patients via examination by a study investigator (n=14) or by contact with local physicians (n=2). TPE was undertaken with continuous-flow centrifugation in all patients. TPE response was assessed prospectively in four patients included in a prior double-blinded, sham-controlled trial[3] and retrospectively in 15 (11 via neurological examination done by a study investigator; four by chart review and physician contact). The response was assessed independently from, and blinded to, immunopathological classification for all patients. Patients were assessed at the Mayo Clinic (n=12), University of Vermont (n=1), or a European centre (n=6). Immunopattern classification was done with 100% consensus among three study investigators, as well as independently from, blinded to, and after clinical assessment and documentation of TPE response for all patients. The figure shows the characteristics of pattern II and III pathology.

The primary outcome was TPE response according to an established grading scale[5] that defines neurological improvement as: none; mild (subjective or minimal, but no effect on function); moderate (gain in neurological status that affects function); or marked (important difference from baseline with major gain in function).

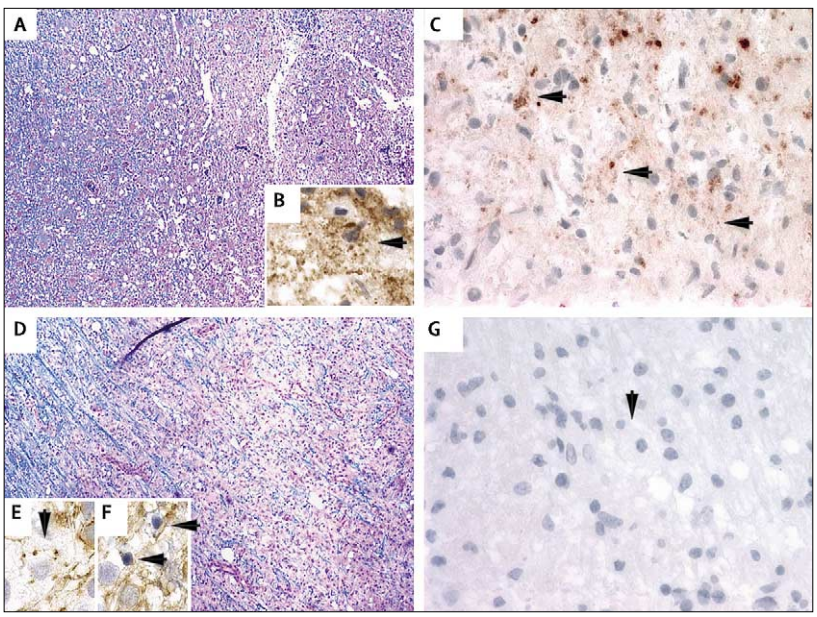

Figure: **Active demyelinating lesions**
Lesions from pattern II TPE responder (A) and pattern III TPE non-responder (D) are hypercellular, with myelin loss (blue on Luxol Fast Blue, Periodic Acid Schiff's stain; ×4) and presence of macrophages containing myelin degradation products (B, E: brown granules in cell cytoplasm labelled with arrows; immunocytochemistry for CNPase; ×600). C9neo deposition, a marker for complement activation, is present within macrophages throughout the lesion in the pattern II TPE responder (C), but not in the pattern III lesion (G: red granules labelled with arrowheads; immunocytochemistry for C9neo; ×400). Oligodendrocyte apoptosis (F: arrowheads; CNPase; ×400) is a hallmark of pattern III, but not pattern I or II, demyelinating lesions.

Improvement was graded for each of the following: cerebral (aphasia, apraxia); motor (hemiparesis, paraparesis, quadriparesis); brainstem or cranial nerve (respiratory failure, diplopia); cerebellar (limb, truncal ataxia); and sensory (sensory useless limb, sensory level) impairment. The outcome was dichotomised as success or failure depending on whether moderate or marked improvement occurred in any neurological deficit observed before TPE. Change in expanded disability status scale score from baseline to 1 month after TPE was used only as a secondary outcome since the scale is insensitive to upper extremity weakness and cognitive or language deficits. Associations between treatment response, immunopathological patterns, expanded disability status scale score, and other demographic or clinical variables were assessed with Fisher's exact or Wilcoxon rank sum tests for categorical or continuous variables, respectively.

	Treatment failure (n=9)	Treatment success (n=10)	Total (n=19)	p*
Multiple sclerosis pathological pattern [n (%)]				<0·0001
Pattern I	3 (33%)	0 (0%)	3 (16%)	
Pattern II	0 (0%)	10 (100%)	10 (53%)	
Pattern III	6 (67%)	0 (0%)	6 (32%)	
Expanded disability status scale [median (range)]†				
Pre-TPE	7·0 (3·0 to 9·5)	7·3 (4·0 to 9·5)	7·3 (3·0 to 9·5)	0·63
1 month post-TPE	6·8 (3·0 to 9·5)	4·0 (2·0 to 8·0)	4·5 (2·0 to 9·5)	0·08
Change pre-to-post	0·0 (-0·5 to 0·0)	−2·0 (−5·5 to 0·0)	−0·75 (-5·5 to 0·0)	<0·0001
Neurological deficit [n/N]‡				
Brainstem/cranial nerve	0/5	4/4	9 (47%)§	0·66
Cerebellar	0/3	1/4	7 (37%)§	1·00
Cerebral impairment	0/9	5/6	15 (79%)§	0·09
Motor weakness	0/5	7/7	12 (63%)§	0·65
Sensory	0/4	4/6	10 (53%)§	0·66

*Fisher's exact or Wilcoxon rank sum tests were used to estimate p values for nominal or continuous variables, respectively. †One pattern III patient lacked the 1-month post-TPE expanded disability status scale estimation and thus was omitted from all median estimates (ie, pre-TPE, post-TPE, and change pre-to-post scores) for both the treatment failure (n=8) and total (n=18) columns. ‡For each specific neurological deficit, n/N represents the number of patients experiencing moderate-to-marked improvement after TPE out of the total number of patients with that deficit. Since most patients had multiple neurological deficits at time of TPE, the numbers exceed the total for each response group (nine for failure, and ten for success). §Values represent number of patients with and frequency (% out of total 19) of each neurological deficit.

Table: **Association between TPE response and pathological pattern of multiple sclerosis, expanded disability status scale score, and neurological deficit**

At the time of TPE, 17 of 19 (90%) patients had multiple neurological deficits. These deficits were due to a first demyelinating event (n=14) or an exacerbation of established relapsing remitting multiple sclerosis (n=5). None had progressive multiple sclerosis. At follow-up, all but one met Poser or McDonald criteria for definite or probable multiple sclerosis. The remaining patient had no recurrence at 20 months' follow-up. All but one patient received high-dose corticosteroids (methylprednisolone >7 mg/kg intravenously or equivalent for >5 days) within 3 months before TPE and were regarded as steroid-unresponsive because of no change in neurological status. One patient received concomitant immunomodulatory treatment within 3 months before TPE. Patients had a median of six exchanges (range 4–7), with the regimen being determined by the treating physician. Immunopattern distribution for pattern I (n=3), pattern II (n=10), and pattern III (n=6) was similar to that of a large multiple sclerosis biopsy cohort.[1] Patterns did not seem to change after TPE since two patients with both biopsy and autopsy had identical patterns before and after TPE (pattern II, n=1; pattern III, n=1).

After TPE, all patients with pattern II pathology (ten of ten), but none with pattern I (none of three) or pattern III (none of six), experienced moderate-to-marked improvement (ie, treatment success) in one or more neurological deficit (table, $p<0·0001$). As previously reported, patients who successfully responded began to improve early after the start of TPE (median 3·0 days [range 1–8]).[4] Whereas baseline expanded disability status scale score before the start of TPE did not differ between those who were treatment successes compared with those who were treatment failures, significant improvement in scores within 1 month after TPE occurred only in those classified as treatment successes (table, $p<0·0001$). However, expanded disability status scale score at last follow-up (2·5 years [0·4–13·9]) did not differ between response groups, consistent with the lack of established long-term efficacy of TPE in reducing relapse frequency or neurological progression in patients with multiple sclerosis.

Sex, age at pathological diagnosis, time from index attack to TPE, and number of exchanges did not differ between those with treatment success or failure. Immunopathological pattern was not significantly associated with any category of neurological deficit. Although a trend for increased frequency of cerebral deficits was recorded in the treatment failure group (p=0·087), none of nine in the failure group compared with five of six in the success group experienced moderate-to-marked improvement in cerebral function. This finding suggests that the presence of cerebral impairment does not preclude a favourable response to TPE. Despite a trend for increased age at time of TPE in the success group relative to the failure group (36·8 *vs* 26·5 years, respectively; p=0·066), a large retrospective TPE study did not report this association.[4]

Our study shows that only patients with pattern II multiple sclerosis pathology, uniquely characterised by immunoglobulin deposition and complement activation, respond to TPE given for fulminant demyelinating attacks. This selective response could indicate a mechanism of action of TPE, namely the removal of pathogenic humoral and plasma factors. T-cell and macrophage inflammation is similar across all three patterns, which suggests that cellular components probably do not account for the recorded differences in TPE response. Improvement after TPE is typically rapid and all-or-none.[3,4] The clear dichotomy between treatment success and failure allows early and reliable clinical identification of responders, even with small sample sizes.

A pronounced TPE response is seen in other presumed humorally mediated disorders, such as neuromyelitis optica.[4] In particular, neuromyelitis optica lesions show perivascular immunoglobulin deposition and complement activation, and such patients have a circulating immunoglobulin G autoantibody specific to this disease.[6] Since TPE non-selectively removes intravascular plasma proteins, the precise mechanism by which TPE improves fulminant CNS IIDD attacks is unknown. In addition to immunoglobulins, complement factors, cytokines, and immune complexes could contribute to humorally associated demyelination. Recent studies suggest antibodies to myelin antigens might be important biological markers of multiple sclerosis.[7] Given the retrospective nature of this study, no sera or cerebrospinal fluid samples were available immediately before and after TPE to analyse reactivity towards myelin proteins, complement factors, classic acute phase proteins, or immunoglobulin classes.

Study limitations include small sample size and retrospective design. The design issue, however, was addressed by strict and blinded classification of immunopattern and clinical outcome measures. Despite our small sample size, the association between TPE response and pattern II patients is robust. Although all pattern II patients showed moderate-to-marked improvement, all patients are not anticipated to respond to TPE in a larger cohort. If severe axonal damage accompanies demyelination, substantial neurological improvement is unlikely given TPE failure in patients with hypotonia and absent muscle stretch reflexes.[4] Although multiple sclerosis biopsy cohorts are susceptible to selection bias, long-term clinical follow-up on a large series (n=91) revealed that most patients develop typical multiple sclerosis, and demographically and clinically resemble a population-based multiple sclerosis cohort.[8]

Our results suggest that pathological differences between multiple sclerosis lesions could account for the variable response to TPE. Serological, genetic, or radiological surrogates need to be identified to reliably differentiate pathological patterns without biopsies to directly apply these findings to clinical practice. Ongoing studies are investigating MRI signatures potentially associated with a favourable TPE response. Whether responses to other multiple sclerosis treatments are associated with distinct pathological subtypes also needs to be addressed.

Contributors

M Keegan was responsible for the design of the study, blinded assignment of clinical outcomes, and manuscript preparation. F König did blinded clinical evaluation of European patients and revised the manuscript. R McClelland did statistical analyses and manuscript preparation. W Brück identified eligible European patients, did blinded pathological analyses and immunopathological classification, and revised the manuscript. Y Morales did statistical analyses and manuscript preparation. A Bitsch contributed to blinded clinical evaluation of European patients and revision of the manuscript. H Panitch did patient evaluations and revised the manuscript. H Lassmann undertook blinded pathological analyses and immunopathological classification, and revised the manuscript. B Weinshenker was responsible for recruitment and blinded clinical evaluation of patients, and revision of the manuscript. M Rodriguez did recruitment and blinded clinical evaluation of patients, and revised the manuscript; J Parisi undertook pathological analysis and revision of the manuscript. C F Lucchinetti directed the study, identified eligible US patients, did blinded pathological analyses, immunopathological classification, and blinded clinical assessment, and followed up patients, interpreted data, did statistical analyses, and prepared the manuscript.

Conflict of interest statement

We declare that we have no conflict of interest.

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