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EDITORIAL REVIEW

Immunopathogenesis of multiple sclerosis: MBP and beyond

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The past decade has witnessed a profusion of studies on the human T cell response to myelin basic protein (MBP). In fact, MBP can be considered by far the best characterized human autoantigen; however, the response that it elicits from the T-cell remains bewilderingly complex: not only do T cells recognize a large number of MBP-epitopes, they also use different HLA-molecules as restriction elements, and use a diverse spectrum of T-cell receptors (TCR). Even less clear is the role of MBP as a potential target antigen in multiple sclerosis (MS). The study by Massa *et al.* [1] addresses this topic in the current issue of *Clinical and Experimental Immunology*. The authors examined T cells from MS patients and controls three times during an 18-month period. With a kinetic response assay, they showed that a large proportion of these patients had a significant response to eight different MBP regions, but the T-cell recognition of the MBP peptides fluctuated – now it appeared, then disappeared, only to reappear later [1]. The importance of these results can be understood only in the context of the following observations and established facts.

IMMUNODOMINANCE AND PEPTIDE BINDING

The region that spans amino acids 80–100 of human MBP is generally considered to be immunodominant, but some reports have shown that the N-terminal or C-terminal parts of MBP can also be immunodominant in individual patients [2–5]. Since HLA-DR2 is associated with MS in Caucasians (about 2/3 of the patients vs. 1/4 of the general population are HLA-DR2+), research has focused on DR2+ subjects. The HLA-DR2 haplotypes contain two nonallelic genes; in Caucasians these are typically HLA-DRB1*1501 (formerly DR2b) and HLA-DRB5*0101 (DR2a). MBP peptides spanning the whole molecule could be presented by different HLA-DRB1 alleles [6]. Specifically, the peptide MBP84-103 bound both gene products of the DR2 haplotype. This agrees with findings that peptides from this region of MBP are presented by both DRB1*1501 and DRB5*0101 transfected L-cells [3,7]. The promiscuous binding of MBP80-99 to different HLA-DRB1 products is the main reason for its immunodominance. Furthermore, MBP80-99 can also be presented by the so-called supertypic HLA-DRB3 gene products (DRw52) [3], which are components of different HLA-DR haplotypes.

Successful crystallization of this complex has provided insight into how MBP85-99 binds to HLA-DRA/B1*1501 [8]. Amino acid residues pointing toward the TCR or the peptide-binding groove of HLA-DR2 have been identified [9,10]. On the basis of structural requirements [11] and with combinatorial peptide libraries [12], human T cells specific for MBP80-100 have been shown to have a number of cross-reactivities to microbial proteins, for example, to peptides encoded by adenovirus type 12 and Epstein-Barr virus.

THE RELEVANCE OF MBP-SPECIFIC T CELLS FOR MS AND THEIR POTENTIAL ENCEPHALITOGENICITY

MBP-specific T cells are present in the circulation of both healthy persons and MS patients [13]. The estimated frequency of MBP-specific T cells depends to a large extent on the method applied to determine it. For example, estimations based on the cloning of T cells as opposed to ELISPOT analysis seem to underestimate the actual frequency [14]. However, when only the pure frequency of MBP-specific T cells in the blood of MS patients and controls was compared, there were no consistent differences, regardless of the approach used.

Further, the diverse character of epitope specificity and TCR repertoire does not seem to consistently differ in MS patients and healthy subjects. Serial (longitudinal) analysis of the MBP-specific T-cell response indicated that there is some degree of clonal persistence and activation of MBP-specific T cells in MS patients [3,15], but this is also true for healthy donors [16]. However, certain functional properties of the MBP-specific T cells differ, e.g. IL-2 responsiveness [14,17], HPRT resistance [18], costimulation requirements [19–21], and IL-7 sensitivity [22]. These studies also indicated that MBP-specific T cells might have been activated *in vivo* in MS patients.

Are the MBP-specific T cells which can be isolated from MS-patients and healthy donors potentially encephalitogenic? The most straightforward way to answer this question would be to directly transfer human MBP-specific T cells into appropriate host animals, either HLA-transgenic mice or MHC-compatible

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monkeys. We showed that some human MBP-specific T cells can recognize their antigen when presented by PBMC from MHC-compatible rhesus monkeys, but surprisingly not with APC from HLA-DR transgenic mice [23]. Thus far, there is no direct functional proof that human MBP-specific T cells are actually encephalitogenic, but a number of different experiments strongly suggest that human MBP-specific T cells are potentially encephalitogenic. For example, autoimmune encephalomyelitis could be induced in humanized mice, which were transgenic for HLA-DRA*0101/DRB1*1501, CD4, and the TCR of an MBP-specific T cell clone [24]. MBP-specific T cell lines have been established from unprimed monkeys and injected into autologous monkeys [25] or a bone marrow chimera [26], providing formal proof that MBP-specific encephalitogenic T cells are present in the circulation of healthy primates.

Perhaps the strongest evidence that MBP-specific T cells are potentially encephalitogenic in humans derives from the adverse effects observed in a recent clinical trial [27], in which an altered peptide ligand (APL) based on MBP peptide (83–99) was used to treat MS patients. During the therapy several patients developed a T-cell response that was cross-reactive with the APL and MBP, and simultaneously had a flare-up of clinical activity [27]. Positive immunohistochemical staining of microglial cells in MS lesions, with a mAb that specifically recognized the complex of DRB1*1501 and MBP peptide 85–99 [28], also suggests that MBP may act as a disease-relevant autoantigen.

In addition to the well-characterized HLA-DR-restricted CD4+ T cells, CD8+ MBP-specific T cells were detected after stimulation with MBP-peptides carrying an HLA-A2-binding motif [29]. Recent demonstrations of myelin-specific CD8+ encephalitogenic T cells [30,31], and expansions of CD8+ T cells in MS lesions strongly indicate that CD8+ cells may be important in MS [32].

ANTIGEN-SPECIFIC TREATMENT FOR MS

On the basis of the above findings and observations in animal models of MS [33], a number of clinical trials attempted to specifically modulate the MBP-specific T cell response in MS patients [34]. T-cell vaccinations containing MBP-specific T cell clones or peptides of selected TCR-Vβ elements were used to induce an anticlonotypic or an anti-TCRVβ specific immune response [35,36]. Variants of MBP83-99, which retained HLA-DR2 binding, were used as altered peptide ligands in the hope of inducing a Th2 shift, an effect that may critically depend on the applied dosage [27,37]. Anergy could be induced in human MBP-specific T cells with dimeric TCR ligands [38]. A complex of DR2 with the MBP-peptide 84–102 is currently being tested in a clinical trial [39].

Despite the impressive advances made in our understanding of MBP-specific T cell responses at the molecular level, there is still considerable skepticism as to the feasibility of such antigen-specific therapeutic strategies. Moreover, many complicating aspects and issues in MS are still emerging; some are summarized in Table 1. In particular, there is increasing evidence that MS is probably not a single disease but rather a heterogeneous group of clinically related disorders [40]. Furthermore, autoimmune reactions are not necessarily always detrimental; they may have benefits as well [41–43]. All this, plus the complexity and fluctuating nature of the anti-MBP immune response, most recently proven by Mazza *et al*. [1], has dampened any over-enthusiastic expectations. The development of 'antigen-specific' MS therapies promises to remain a formidable challenge for some time.

Table 1. Various aspects of autoimmune responses in MS that complicate the development of 'antigen-specific' therapies

(1)	Microheterogeneity of T-cell recognition: one peptide might act as an APL for one T-cell clone, while at the same time be fully stimulatory for another
(2)	Diversity of TCRs recognizing the same DR2-peptide complex
(3)	Different immunodominant regions of MBP in different patients
(4)	Pathogenic role of other autoantigens (MOG, PLP, S-100β, MOBP, alpha B crystalline, CNPase, etc.)
(5)	Role of CD8 + encephalitogenic T cells
(6)	Disease heterogeneity of MS
(7)	(Neuro-)protective role of CNS autoimmunity

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