

Multiple approaches to multiple sclerosis

Multiple sclerosis results from the failure of several different regulatory mechanisms designed to protect against autoimmunity, suggesting multiple targets for therapeutic intervention. Three papers in this issue suggest that if tolerance to components of the nervous system is not maintained in the thymus and autoimmunity ensues, the extent of brain damage can be checked by blockade of glutamate receptors on neurons and oligodendrocytes (pages 56–70).

MULTIPLE SCLEROSIS (MS), an inflammatory disease that affects nearly one million people worldwide, arises when the immune system mistakenly attacks self molecules within the white matter of the brain and spinal cord¹. The immune system ordinarily uses several mechanisms to prevent the development of autoimmune responses against the brain. Indeed, the thymus expresses molecules, including myelin proteins, which are involved in the insulation of axons within the nervous system. Thymic expression of these molecules helps to maintain immunological tolerance through 'education' of the developing immune system. This education enables T cells to distinguish self molecules from foreign invaders. In MS, the regulatory mechanisms that guard against autoimmunity are bypassed, and inflammation in the central nervous system results.

In this issue of *Nature Medicine*, three reports define 'checkpoints' involved in determining the onset of autoimmunity in the nervous system. The checkpoints have three distinct locations: A report from Klein *et al.* suggests that essential events in the development of MS occur in the thymus², where the T-cell repertoire learns to distinguish self from non-self. Pitt *et al.* and Smith *et al.* provide data that indicate that checkpoints also occur in the white matter of the central nervous system, where oligodendrocytes are damaged by the inflammatory attack, and the gray matter, where neurons undergo a secondary degeneration resulting from the loss of white matter^{3,4}. Many of the main structural components of the myelin sheath, including myelin basic protein and proteolipid protein (PLP), are expressed in the thymus⁵.

LAWRENCE STEINMAN

'Ectopic' or 'promiscuous' expression of these molecules within the thymus causes negative selection of developing T cells capable of reacting with PLP, and deletion of these potentially autoreactive T cells. This is a chief factor in the maintenance of self tolerance and the preven-

tion of autoimmunity. PLP, one of the main components of the myelin sheath, exists in two isoforms as a result of alternative mRNA splicing: a longer form, called PLP, and a shorter isoform, called DM20, which lacks a loop of 35 amino acids. The DM20 isoform is expressed on thymic epithelium in mice that are both resistant and susceptible to experimental autoimmune encephalomyelitis (EAE). Mice that are resistant to EAE do not express MHC class II molecules capable of presenting peptides derived from the PLP

35 amino-acid loop, which is found in the central nervous system. Susceptible strains, on the other hand, express MHC class II molecules that allow presentation of peptides from this 35 amino-acid loop to T cells. When these mice are immunized with either peptide fragments from this loop or native PLP, they develop vigorous T-cell responses to PLP and become paralyzed. These findings may help explain why genes in the class II region of the MHC are the leading factor in genetic susceptibility to MS^{6,7,8}.

Klein *et al.* suggest that a fragment of a myelin gene may be expressed in human thymus, and that individuals with a particular MHC genotype may be capable of presenting brain-specific fragments of myelin to un-tolerized T cells². This may lead to the development of demyelinating disease. There are many strategies for re-tolerizing an individual with demyelinating disease once the thymic checkpoint has been evaded and autoimmunity is ongoing^{1,9}. Some of these strategies, such as the use of Copaxone, involve peptide-based drugs that block MHC presentation of brain specific myelin fragments and have been approved for treat-

ment of MS. Other experimental peptide-based drugs are in clinical trials.

Two other checkpoints that determine prevention or susceptibility to EAE also exist, and involve classical neuroprotection strategies (Fig. 1). Neurodegeneration in the brain can be regulated by a class of neuroprotective compounds that interfere with receptors for the excitatory neurotransmitter glutamate. Inhibition of glutamate receptors has been a popular strategy to mitigate damage in gray matter diseases

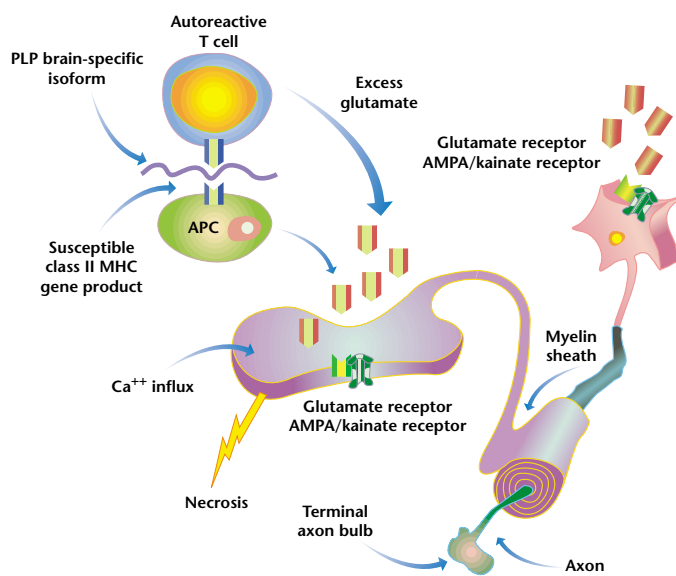


Fig. 1 Possible role of AMPA/kainate receptors on neurons and glia. Activation of T lymphocytes (in this case a T lymphocyte reactive to the brain-specific loop of the myelin protein PLP), macrophages and resident microglia at the site of inflammation in the white matter of the central nervous system causes release of glutamate. The increased extracellular glutamate binds to AMPA/kainate receptors on neurons and on oligodendrocytes. This leads to increased calcium fluxes and death of oligodendrocytes and neurons.

Bob Crimi

of neurons, such as stroke, epilepsy, and neurodegenerative conditions like Huntington disease or Parkinson disease. Certain types of glutamate receptors are found on both neurons and on the glial cells that make myelin, called oligodendrocytes. Blockade of these receptors has been found effective in suppressing damage in inflammatory disease within the white matter of the brain in EAE (ref. 9), the leading animal model of MS.

AMPA (α -amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid)/kainate receptors, which mediate toxicity induced by the excitatory neurotransmitter glutamate, are present on oligodendrocytes and neurons. During inflammation in both EAE and MS, lymphocytes, brain microglia and macrophages release excessive amounts of glutamate, which can then activate AMPA receptors. Pitt *et al.* and Smith *et al.* have shown that antagonists of these receptors can ameliorate EAE in rodents, and prevent clinical relapses that occur when treatment is begun after the onset of paralysis^{3,4}. The blockade of AMPA/kainate receptors does not influence the immune response to myelin antigens, but somehow protects oligodendrocytes from immune-mediated damage. Damage may be mediated by increased calcium flux, which may cause necrotic damage to oligodendro-

cytes. Axonal transection is an essential component of the pathology of MS and EAE (refs. 9,10), and axonal damage is also reduced when AMPA/kainate antagonists are used to treat EAE (ref. 3). The use of neuroprotective agents that block sub-types of glutamate receptors has been an important factor in the development of therapies for stroke and neurodegenerative conditions. The findings of Pitt *et al.* and Smith *et al.* indicate that neuroprotective agents designed to block glutamate receptors may also be useful for treatment of immune-mediated diseases of the gray and white matter in the central nervous system.

It may be possible, through early intervention, to block an immune response to a peptide fragment of a myelin antigen that escaped thymic tolerization, medicating with drugs like Copaxone or altered peptide ligands^{1,9}. Later in the disease process, it may be possible to augment this specific form of immune suppression with neuroprotective agents that block glutamate receptors, and protect both oligodendrocytes and axons from necrotic damage. Taking these concepts from the bench to the bedside may prove beneficial to individuals suffering from MS, both in its early stages and in its more chronic forms.

1. Steinman, L. Multiple sclerosis: A coordinated immunological attack against myelin in the central

nervous system. *Cell* **85**, 299–302 (1996).

2. Klein, L., Klugmann, M., Nave, K., Tuohy, V. & Kyewski B. Shaping of the autoreactive T-cell repertoire by a splice variant of self protein expressed in thymic epithelial cells. *Nature Med.* **6**, 56–61 (2000).
3. Pitt D., Werner, P. & Raine, C. Glutamate excitotoxicity as a mechanism in multiple sclerosis. *Nature Med.* **6**, 67–70 (2000).
4. Smith, T., Groom, A., Zhu, B., & Turski, L. Autoimmune encephalomyelitis ameliorated by AMPA antagonists. *Nature Med.* **6**, 62–66 (2000).
5. Pribyl, T.M., Campagnoni, C., Kampf, K., Handley, V.W. & Campagnoni, A.T. The major myelin protein genes are expressed in the human thymus. *J. Neurosci. Res.* **45**, 812–819 (1996).
6. Haines, J.L. *et al.* A complete genomic screen for multiple sclerosis underscores a role for the major histocompatibility complex. The Multiple Sclerosis Genetics Group. *Nature Genet.* **13**, 469–471 (1996).
7. Ebers, G.C. *et al.* A full genome search in multiple sclerosis [see comments]. *Nature Genet.* **13**, 472–476 (1996).
8. Sawcer, S. *et al.* A genome screen in multiple sclerosis reveals susceptibility loci on chromosome 6p21 and 17q22 [see comments]. *Nat. Genet.* **13**, 464–468 (1996).
9. Steinman, L. Assessment of the utility of animal models for multiple sclerosis and demyelinating disease in the design of rational therapy. *Neuron* **24**, 511–514 (1999).
10. Trapp, B.D. *et al.* Axonal transection in the lesions of multiple sclerosis. *N. Eng. J. Med.* **338**, 278–285 (1998).

Dept. of Neurology and
Neurological Sciences
Beckman Center for Molecular Medicine
B002
Stanford University
Stanford, California 94305, USA
Email: steinman@stanford.edu

Germ cell transplantation—a fertile field

Spermatogonial cell transplantation from a mouse with a defective soma to a mouse with a compromised germ line re-establishes spermatogenesis. The ability of both of these cell types to resume normal function has implications for fertility treatment (pages 29–34).

GAMETOGENESIS IS A complex process that involves the cooperation of two distinct cellular compartments: the germline and the soma (Fig. 1). Not surprisingly, it is prone to errors, and defective sperm production is believed to be responsible for one-third to one-half of all infertility cases. Although most types of male infertility remain idiopathic, developments in micro-assisted fertilization, principally intracytoplasmic sperm injection (ICSI), have substantially improved the chances of reproduction for the one of six couples that experience infertility. In some countries, almost one percent of births result from assisted reproductive therapies (ART). Many of these techniques are dependent on the recovery of hap-

loid cells from the testis, and are usually limited to the use of spermatozoa. In this issue of *Nature Medicine*, Ogawa *et al.* report findings that suggest new therapeutic approaches for male infertility¹.

Spermatogenesis, the process by which a diploid spermatogonial stem cell differentiates into a mature, haploid spermatozoa, occurs within the seminiferous epithelium (Fig. 1). This multi-step pathway begins with mitosis of spermatogonial stem cells². Differentiating spermatogonia enter a protracted meiotic prophase and after the two meiotic

divisions form haploid round spermatozoa. Extensive cellular remodeling and genome repackaging follow to produce mature spermatozoa. Germ cell development occurs in close association with the Sertoli cells, which provide them with structural support, nutrients and regulatory/paracrine factors; the role(s) of the last are still poorly understood because of our inability to reconstitute spermatogenesis *in vitro*. Brinster and colleagues have made important advances in describing the interaction between the germ cells and Sertoli cells within the seminiferous epithelium (Fig. 1) (ref. 3). In this issue of *Nature Medicine*, they report that spermatogenesis can be restored in infertile testis through germ cell transplantation¹.

HOWARD J. COOKE¹ &
PHILIPPA T.K. SAUNDERS²