Experimental autoimmune uveoretinitis (EAU) versus experimental allergic encephalomyelitis (EAE): a comparison of T cell-mediated mechanisms

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SUMMARY

EAU is a model of ocular inflammatory disease. EAU resembles another T cell-mediated autoimmune disease—experimental allergic encephalomyelitis—since both have increased expression of MHC class II molecules in the target tissue, can be adoptively transferred by activated CD4+ T cells and are inhibited by cyclosporin A. The immunological findings will be compared to find out if the same cellular mechanisms are involved in both diseases.

Keywords EAU EAE immune-privileged site autoreactivity

INTRODUCTION

EAU in the rat is a good experimental model for posterior uveitis in man due to the similarities both in the pathology of the disease processes and in the function of the blood-retinal barriers in rat and man [1]. In EAU, infiltrating lymphocytes (mainly CD4+ T cells) appear within the retina early in the disease, resulting in the irreversible destruction of the photoreceptor cells and a loss of integrity of the retinal layers. Throughout the disease there is an increased expression of MHC class II molecules on a variety of retinal cells. At later stages of disease increased numbers of CD8+ T cells can be seen within the retina and it has been proposed that these cells could down-regulate the disease process [2]. The importance of CD4+ T cells in EAU was first demonstrated in the Lewis rat where disease was successfully induced in naive recipients by adoptively transferring activated antigen-specific CD4+ T cell lines [3]. Both cyclosporin A (CsA) and FK506 have been shown to be effective in preventing EAU, suggesting a pathogenic role for activated T cells in this disease [4,5]. EAU has therefore been used as a model to determine the likely T cell-mediated effector mechanisms within the retina. The aim of this review is to compare the findings obtained in EAU with those from EAE, another model of T cell-mediated autoimmunity, to find out if these diseases, both occurring in so-called 'immunologically-privileged sites', have similar effector T cell mechanisms.

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INDUCTION

EAU is induced by immunization with purified retinal proteins or their peptide fragments emulsified in Freund's complete adjuvant (FCA). The most commonly used uveitogen is retinal soluble antigen (SAg), a 48-kD intracellular protein which exists in rod outer segments and the pineal gland [6]. However, two other purified proteins—interphotoreceptor retinoid binding protein (IRBP) and rhodopsin—have also been found to be uveitogenic in some species [7,8]. Synthetic peptides made from these molecules can be uveitogenic, and studies of the EAU induced by SAg peptides have allowed identification of highly pathogenic epitopes within the whole antigen [9]. More recent studies with IRBP have found epitopes of the molecule which are highly uveitogenic but not necessarily stimulatory for T cells in vitro [10]. It has also been found that uveitopathogenic regions of SAg share sequence homology with a variety of viral peptides [11] and EAU has been induced by immunizing with a small synthetic peptide corresponding to amino acid positions 106-121 of yeast histone H3 [12]. Thus molecular mimicry has been proposed as a mechanism for initiating and/or perpetuating EAU.

Chronic relapsing EAE was initially described in guinea pigs using spinal cord homogenate emulsified in FCA [13] but disease has subsequently been induced in a range of species with myelin basic protein (MBP) extracted from white matter. Studies using synthesised peptides from MBP have revealed encephalitogenic sites on the molecule [14]. Recently other central nervous system (CNS) antigens have also been shown to be encephalitogenic, e.g. proteolipid protein [15]. Molecular mimicry has been implicated in EAE due to the finding that many viral infections including measles and the coronavirus JHM can increase...
sensitivity to MBP [16,17] and homologous sequences have been identified between viral peptides and MBP [18].

Alternative methods for inducing EAE and EAU have been demonstrated using adoptive transfer techniques, by immunizing a sufficient number of activated antigen-specific, CD4+ T cells. In early studies induction of EAU by adoptive transfer utilized lymph node cells from SAg-primed rats which initiated a uveitis and associated pinealitis [19]. The induction of EAE in rats by this method was demonstrated using lymph node cells reactive to MBP which induced a chronic demyelinating disease [20]. Exactly how these CD4+ T cells, when injected peripherally, induce an organ-specific disease remains unclear. Studies using radioisotope labelling to follow the migration of the T cells after adoptive transfer of EAU in Lewis rats found only very few of the labelled cells crossed the blood-retinal barrier (BRB) into the eye [21]. This technique had previously been used in EAE to examine the trafficking of lymphocytes into the CNS across the blood–brain barrier (BBB) and an increased accumulation of cells was detected within the CNS before onset of disease [22]. Thus a clonal expansion of the inoculated autoreactive T cells within the tissue has been suggested as the amplification mechanism whereby very few cells can induce EAE or EAU in syngeneic hosts [23,24]. By comparing the methods for inducing EAU and EAE, it appears that in both models similar immunological mechanisms are involved and molecular mimicry has been implicated due to the regions of homology shared between the relevant autoantigens and various peptides.

GENETIC PREDISPOSITION

Models of EAU have been developed in guinea pigs, rats, rabbits and mice although the uveitogenic responses within species are varied with some strains of rats (PVG and Lewis) being more susceptible to disease than others [24]. In mice, EAU has only recently been achieved, and its induction requires the use of IRBP as opposed to SAg which appears to be poorly uveitogenic in this species [25]. In EAE, many species are susceptible to disease with some rat (Lewis) and mouse (SJL, PL/J) strains being more susceptible than others [26]. Several studies have attempted to map these responses to the MHC class II gene and, in the mouse, have found that a genetic predisposition to EAE partly depends on the s and q haplotypes [27]. In rats, the Lewis strain is susceptible to EAE (RT-I' locus) whereas the BN strain is resistant (RT-1q) suggesting that the MHC class II gene controls the T cell responses to MBP and hence disease susceptibility [28]. However, PVG rats which are relatively resistant to EAE respond to the same MBP peptide as the susceptible Lewis rats, suggesting that the MHC class II gene is not the only factor involved in disease susceptibility [29]. In conclusion, on comparing EAU with EAE, there are many species which are susceptible to disease although the requirements for genetic predisposition remain unclear and there is no evidence that these are the same in both diseases since it is clear that whilst PVG rats are highly susceptible to EAU, this is not true for EAE.

IMMUNOPATHOGENESIS

The immunopathology of EAU is similar to that of EAE. At the early stage of disease within the retina in EAU, large numbers of infiltrating CD4+ T cells and an increase in local expression of class II molecules can be seen [2]. In EAE, infiltrating perivascular CD4+ T cells and MHC class II-expressing cells predominate in the lesions within the white matter of the central nervous system, suggesting that the CD4+ T cells are mediating the demyelination [26].

The various effector T cell mechanisms which might be occurring during EAE and EAU have been examined in vivo using MoAbs recognizing T cell subsets and lymphokines in an attempt to alter the course of disease. Thus EAE in mice has been prevented using either anti-CD4 or anti-class II MoAbs [30,31]. Similarly in EAU, uveitis in rats has been inhibited with anti-I-A or anti-CD4 MoAbs [32,33]. In contrast, no change was seen in the course of disease when Lewis rats were depleted of their CD8+ T cells before inducing EAE although CD8+ T cells have been identified at the later stages of disease [34]. This suggests that these cells do not play an important role in initiating or down-regulating EAE. Similar studies treating rats with anti-CD8 MoAb before inducing EAE failed to alter the course of disease, suggesting that CD8+ T cells are not important in regulating EAU [35].

Anti-lymphokine antibodies can also prevent disease and a recent report of the successful prevention of EAU in Lewis rats with anti-interferon-gamma (IFN-γ) antibody demonstrates the importance of this lymphokine in the pathogenesis of EAU [36]. In Lewis rat EAE, it has recently been shown that IL-1α treatment exacerbates disease and suppression of disease can be obtained by treatment with soluble IL-1 receptor [37]. Another lymphokine which has been found to play an important role in down-regulating chronic relapsing EAE in mice is transforming growth factor-β [38]. Although its role in EAU is unclear, it has recently been detected in high concentrations in normal aqueous humor from several different mammalian species [39].

IMMUNOTHERAPY

Following the success of the adoptive transfer method for inducing EAE with activated CD4+ T cells, it was demonstrated that the same T cells, when inoculated into rats in subencephalitogenic doses, were able to confer protection against subsequent attacks of EAE [40]. This approach has also been successful in vaccinating rats against EAU [41]. The mechanism whereby T cell vaccination prevents disease is thought to occur by initiating an immune response against endogenous clones of lymphocytes with anti-MBP receptors or by enhancing suppression. An alternative method for down-regulating disease has been described in EAU, using a CD8+ 'suppressor' T cell line [42]. One effective immunotherapy in EAE has been peptide-specific prevention of disease. By neonatally tolerizing with immunodominant peptides of MBP, adult mice were subsequently made resistant to EAE [43]. It is not known if this form of immunotherapy is effective in EAU. Another successful approach has been to immunize with peptides selected on the basis of their ability to bind class II molecules and so prevent EAE by blocking class II-mediated processes [44]. Molecular studies of the DNA rearrangements of the T cell receptor variable (V) genes have indicated that encephalitogenic T cell lines share a high degree of homology [45]. This suggests that the T cell response in EAE is polyclonal and that one form of immunotherapy might be to use peptides which can bind to and block antigen recognition sites on T cell receptors and so prevent disease. Using this approach, it has been reported that rats can
be rendered resistant to the induction of EAE by vaccination with a synthetic peptide representing a hypervariable region of the T cell receptor (TCR) Vβ8 molecule [46]. Uveitogenic T cell lines have been examined for their T cell receptor V genes and it has been found that these cells are enriched for Vβ 8.2 [47]. Whether peptides synthesised from this T cell receptor V gene can protect rats from EAU has yet to be demonstrated. Interestingly, oral immunization has been shown to be effective in preventing both EAE and EAU [48,49]. Feeding rats with MBP or SAg resulted in a markedly diminished encephalitis or uveitis. The precise mechanisms involved in this immunosuppression remain unclear although the effects could be reversed by treating the rats with anti-CD8 MoAb. In conclusion, of those immunotherapeutic approaches which have been performed in both EAU and EAE, no immunological differences in the results could be found. It is therefore not surprising that both immunosuppressive drugs CsA and FK506 are effective in preventing EAE and EAU [4,5].

REVERSIBILITY OF DISEASE
In EAE, the perivascular inflammatory infiltrate leads to damage of the myelin sheath surrounding the nerve although the myelin-producing oligodendrocytes can remyelinate [50]. In contrast in EAU, the damage to the photoreceptor cells is irreversible as these cells are not regenerated. Despite this difference in the course of disease, neither photoreceptor cells nor oligodendrocytes express class II molecules in vivo or in vitro. This seems to contradict the theory of aberrant expression of class II molecules on the target cells leading to a perpetuation of chronic inflammatory disease [51]. Perhaps the oligodendrocytes and photoreceptor cells are damaged as a bystander effect during the effector T cell response. The MHC class II molecules, necessary for recognition by CD4+ T cells, have been detected within the retina during EAU on retinal endothelial cells [52] and on pigment epithelial cells [53] although this finding has been disputed by others [54]. In EAE, class II expression has been seen within the lesions, particularly at the perivascular cell level [55]. Class II induction has also been demonstrated in vitro on brain-derived endothelial cells, microglia and both types I and 2 astrocytes but not on oligodendrocytes [56–59], suggesting that a lack of expression of class II molecules by the target cells themselves does not necessarily prevent them being damaged during a T cell response.

BLOOD–BRAIN BARRIER VERSUS BLOOD–RETINAL BARRIER
In both EAE and EAU, successful adoptive transfer of disease requires that the T cells are activated and it has been found that activated T cells produce enzymes capable of degrading extracellular matrix [60]. It has also been recently shown that any activated T cell can cross the BBB [61] and, presumably, the BRB. Exactly how the CD4+ T cells cross the BBB and BRB from the blood remains unclear, but adhesion molecules are thought to be important. An upregulation of adhesion molecules has been reported within the CNS coinciding with the onset of relapsing EAE which supports a role for receptor-mediated migration of cells into the brain [62]. Cultures of cerebrovascular endothelial cells from Lewis rats have been shown to be more adhesive for activated than for resting lymphocytes and this is thought to involve the CD11a/18 complex of adhesion molecules since adhesion could be selectively inhibited by anti-LFA-1 antibody [63]. No such studies have been done with rat retinal endothelial cells but both human retinal endothelial cells and human pigment epithelial cells have been found to express high levels of ICAM-1 following incubation with IFN-γ [64].

Although the BBB and BRB appear to have similar functions in rat and man, one important difference is in the ability of CsA to down-regulate disease. In man, CsA is only poorly effective as a treatment for multiple sclerosis and does not enter the brain in detectable amounts [65]. In contrast, CsA is an effective treatment for posterior uveitis in man [66], suggesting either that it exerts its effect at the peripheral level and reduces the numbers of cells recruited to the site of inflammation, or that it crosses the BRB to locally suppress the T cell response. Evidence that low levels of CsA can cross the BRB in uveitis in man supports the latter hypothesis [67].

CONCLUSION
From the studies carried out so far, EAU and EAE seem to involve similar immunological processes for disease induction, the cytokines produced and the appropriate target molecules for immunotherapy. We were unable to identify any immunological differences either in the induction or in the expression of the disease between EAU and EAE. Some differences exist mainly as a result of anatomical differences between the brain and the eye. Since in both models the autoantigens are localized within the target organ, autoreactive T cells could be attracted to the site of inflammation although it is not clear if in either model the autoreactive T cells expand before or after crossing the relevant barrier.

These two models of T cell-mediated autoimmunity share many common immunological features although their relevance to human disease is limited. Evidence that MBP and SAg are the autoantigens in human disease is questionable since lymphocyte responses to these antigens have been detected in the blood from controls. Nevertheless, these models are clearly of great importance in allowing a further understanding of various autoreactive T cell mechanisms.

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