Understanding Autoimmune Uveitis through Animal Models

The Friedenwald Lecture

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For more than 20 years, my research interests have concentrated on the cellular, molecular, and genetic mechanisms involved in the breakdown of self tolerance to immunologically privileged antigens found in the retina of the eye and in the subsequent pathologic processes that ultimately manifest as autoimmune uveitis. Although my work has emphasized autoimmune uveitis, many of the mechanisms are applicable to other ocular inflammatory diseases, as well as to autoimmune and inflammatory diseases affecting other organs.

The eye has a special relationship with the immune system, known as ocular immune privilege, which we believe to have evolved to protect vision from damage caused by day-to-day inflammatory insults.^{1,2} One of the convincing and medically important manifestations of ocular immune privilege is the extraordinary success of corneal grafts.² However, despite ocular immune privilege, we develop autoimmune uveitis. This is seemingly a paradox that should be reconciled with what we know, or what we think we know, about immune privilege of the eye. Autoimmune uveitis in humans is not a single entity, but rather is a group of diseases that can differ in presentation and clinical course. The eye may be the only organ involved, or uveitis may be part of a syndrome involving multiple tissues.³ Lymphocytes from peripheral blood of uveitis patients often respond to antigenic proteins expressed in the retina, and it is felt that these responses play a central role in the pathogenesis of autoimmune uveitic disease. Together, these diseases are estimated to affect 2 million Americans and to underlie at least 10% of the severe visual handicap.⁴ Because the etiology is usually obscure and because studies on clinical material are hampered by strict ethical considerations and limited availability of tissue samples, it is necessary to use animal models to unravel the basic mechanisms of disease.

ANIMAL MODELS OF AUTOIMMUNE UVEITIS: ENTER THE MIGHTY MOUSE

When I started my work in this field, there was an existing model of experimental autoimmune uveitis (EAU) in rats that could be elicited by immunization with retinal arrestin, which at that time was known as retinal soluble antigen, or S-Ag.^{5,6} The rat model was, and still is, very useful for developing and evaluating therapeutics. Our early studies using this model identified a central role for S-Ag-specific effector T lymphocytes in pathogenesis of EAU.⁷ Nevertheless, the usefulness of the rat model for basic immunologic studies was hampered by

limited availability of immunologic reagents and lack of genetically defined and genetically manipulated strains. Many laboratories made efforts to duplicate the model in mice, but unfortunately, the commonly available laboratory strains of mice appeared resistant to disease induction with S-Ag, leading to more frustration than results.

The breakthrough came in 1986, when Igal Gery⁸ and his collaborators identified interphotoreceptor retinoid binding protein (IRBP) as a strong uveitogenic antigen in Lewis rats. IRBP also proved to be pathogenic in several commonly available mouse strains, a finding that we reported in 1988.⁹ Interestingly, several of the mouse strains also developed some disease with S-Ag, but attaining consistent results continued to be a challenge, leaving IRBP as the antigen of choice in the mouse.

The classic EAU model in mice, as reported in 1988 and modified subsequently,^{10,11} is induced with IRBP, a major retinal protein that shuffles vitamin A derivatives between the photoreceptor cells and the retinal pigment epithelium (RPE). For induction of EAU, IRBP is emulsified in complete Freund's adjuvant (CFA), which consists of a suspension of tuberculosis bacteria in mineral oil. CFA is critical for disease induction in both mice and rats; the reason for this will be discussed ahead. Many mouse and rat strains also require an additional inflammatory stimulus in the form of pertussis toxin (PTX). As with humans, where not everyone will develop uveitis, susceptibility in rodents is strain-dependent. In those strains that do develop disease, as well as in rats, the pathology is quite reminiscent of human uveitis, and it is believed that essential cellular mechanisms of disease are shared in rodent and in humans.

The establishment of the IRBP-induced mouse model of EAU literally catapulted forward the basic research into immunologic mechanisms of the disease, such that within a few years, the number of publications using mouse models outnumbered those in the rat (Fig. 1). This was particularly aided by identification of the major pathogenic molecular sequences for strains that demonstrated susceptibility, permitting laboratories that were not equipped to purify native IRBP, to use synthetic peptides representing short fragments of this protein to induce disease.¹²⁻¹⁴ Thanks to the availability of a plethora of immunologic reagents and genetically defined as well as genetically engineered mouse strains, tools have gradually been developed that allow us to ask questions about the genetic and cellular control of uveitic disease at a level of sophistication that was not possible before.

Early studies of susceptibility in which we used congenic mouse strains, which share the MHC region, but differ in their genetic background, or share the same genetic background, but differ in MHC, demonstrated that both MHC and non-MHC genes affect susceptibility and helped to define the strains that are susceptible or resistant to EAU. Thus, the C57Black background (B10 or B6) is permissive to EAU expression, provided

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FIGURE 1. EAU model in rats and mice: annual publications. A PubMed search was performed with keywords that included permutations of “mouse,” “rat,” “EAU,” “experimental autoimmune,” and “uveitis or uveoretinitis” and excluded irrelevant hits and review papers, delimited by calendar year of publication, to yield the number of papers per year. Papers for 2010 were adjusted by fraction of year elapsed at the time of analysis (October 2010).

that there is a susceptible H-2 haplotype such as H-2^r, H-2^k (or the closely related H-2^a) or H-2^b, in that order of susceptibility.¹⁵ The study of human MHC molecules in susceptibility was made possible by adaptation of the EAU model to “humanized” mice, which are HLA-transgenic strains in which mouse MHC molecules have been replaced by human MHC molecules. We showed that several human HLA class II molecules support EAU development in response to S-Ag, which is the main antigen recognized by lymphocytes of uveitis patients, but to which wild-type parental mouse strains are resistant.¹⁶ This humanized model is now making possible the study of the retinal antigens and their fragments that are pathogenic in the context of the human MHC molecules. In parallel, the developing rat genetics and genomics permitted an increasing level of analysis of the contribution of background genes to disease susceptibility, demonstrating that many non-MHC uveitis susceptibility loci are common to other autoimmune diseases and are shared between animals and humans,^{17,18} further supporting the usefulness of these models for the study of basic disease mechanisms.

Other variations on the mouse model of EAU are proving useful for studying cellular mechanisms of autoimmunity to retina. These include EAU induced by peripheral injection of retinal antigen-pulsed dendritic cells (DCs) rather than injection of the antigen emulsified in CFA.¹⁹ This model, recently developed in our laboratory, may help to explain the clinical heterogeneity of uveitic disease (discussed later). A particularly promising model, also recently developed in our laboratory, is a mouse expressing a transgenic a T-cell receptor (TCR) specific to IRBP, which has a much higher than normal frequency of these cells, greatly amplifying the relevant responses. These animals develop spontaneous EAU with high incidence and offer to improve on the hitherto available approach of studying retina-specific T cells in mice expressing a foreign protein as a neoantigen in the retina and a transgenic TCR that recognizes this protein (e.g., hen egg lysozyme, β galactosidase, or influenza hemagglutinin; reviewed in Ref. 20).

AUTOIMMUNE UVEITIS: THE WHY, THE HOW, AND THE WHY NOT—LESSONS FROM ANIMAL MODELS

Using the different variants of the mouse uveitis model and aided by the rat model of uveitis, over the years we and others have been able to define critical checkpoints in the pathogenesis of uveitis, which we believe are also applicable to human

disease (Fig. 2). Although in this review I will emphasize data produced in my own laboratory, I wish to recognize with great appreciation the extensive contributions of scientific colleagues in the United States and abroad, whose work has been invaluable to our understanding of the mechanisms driving uveitis.

To understand autoimmune uveitis, we must understand:

WHY we develop disease, since we are supposedly immunologically tolerant to self, and the eye enjoys an immune privileged status;

HOW the immune system is triggered to react and what events must take place to lead to tissue pathology; and finally,

WHY NOT—that is, what prevents most of us, most of the time, from developing the disease.

As mentioned, autoimmune uveitis induced experimentally is a T-cell-dependent disease in rodents. This is also the case apparently in human disease, as clinical uveitis is ameliorated by T-cell targeting therapies, including cyclosporine, rapamycin, and IL-2 receptor targeting.³ Thus, it is likely that the basic cellular and molecular mechanisms as studied in rodents will be reasonably representative of human disease.

The WHY

Our propensity to develop uveitis is intimately connected to the T lymphocytes that recognize retinal components and have the potential to attack it as though it were foreign tissue, causing autoimmune uveitis. As part of their normal development, T cells emigrate from the bone marrow as immature precursors and enter the thymus where they differentiate and mature, and finally exit into the periphery, ready to do battle with invading pathogens. During their sojourn in the thymus, the maturing T cells undergo a process of “education” to ignore proteins (antigens) that compose our own tissues, whose component proteins are ectopically expressed in the thymus precisely for this purpose. Examples of such tissue-specific self antigens include pancreatic insulin, heart myosin, and retinal antigens, among them IRBP.²¹ The process of thymic education eliminates or renders anergic the T cells that happen to express antigen receptors with high affinity to self, and spares those with low affinity to self for the purpose of host antimicrobial defense. However, as with all biological processes, thymic education is not fully efficient, and self-reactive cells that should have been eliminated exit from the thymus into the periphery. Normally, such cells would have been dealt with by a second tier of control, known as peripheral tolerance, where such “escapee” T cells recirculating through the body come in contact with tissue antigens under noninflammatory conditions and as a result are rendered specifically tolerant of these antigens. However, retinal antigens residing within the healthy eye are largely sequestered behind the blood-retinal barrier as part of ocular immune privilege, hindering the normal functioning of peripheral tolerance. This causes persistence of a population of nontolerant T cells that can be activated by a chance exposure to antigen, either as a result of ocular trauma or in the form of a microbial mimic whose molecular structures happen to resemble self. Several molecular mimics of retinal antigens have been described (reviewed in Ref. 22).

Indeed, the eye seems overwhelmingly dependent on the thymus as the mechanism of self tolerance. Experimental evidence produced by us and by others has shown that mice that fail to eliminate retina-specific T cells in the thymus have enhanced susceptibility to uveitis: (1) Mice implanted with a thymus from an IRBP-knockout donor that lacks expression of IRBP in the thymus; these mice have detectably altered immune responses to IRBP and when immunized with a low dose

FIGURE 2. The Why, How, and Why Not: critical checkpoints in pathogenesis of uveitis. Imperfect negative selection in the thymus results in export to the periphery of retinal antigen-specific T cells. Because of relative sequestration of retinal antigens in the eye, they can be activated by retinal (trauma) or cross-reactive (mimic) antigens in the context of costimulatory “danger” signals, whereupon they escape from control of natural Tregs and differentiate into pathogenic effector T cells. Following clonal expansion, some of them reach the eye. Resisting local regulatory mechanisms, they break down the blood–retinal barrier, activate the retinal vasculature, and recruit inflammatory leukocytes from the circulation. The resulting inflammation causes damage to the tissue and leakage of ocular antigens, activating eye-dependent systemic (and local?) regulatory mechanisms that terminate the disease and limit pathology. mTECs, medullary thymic epithelial cells. Modified from Caspi RR. Ocular autoimmunity: the price of privilege? *Immunol Rev* 213:23–35.

of IRBP, they develop abnormally severe disease.²³ (2) A similar situation exists in mice that are genetically deficient in the transcriptional regulator AIRE, which controls thymic expression of tissue antigens (including IRBP). They too fail to delete retina-specific cells and, over time, develop spontaneous uveitis.²⁴

Conversely, forced expression of unique retinal antigens in the peripheral tissues, in a nonprivileged environment, induces immune tolerance and protects from uveitis. For example, extraocular expression of IRBP can be elicited in normal adult animals by hydrodynamic injection of an IRBP expression plasmid as naked DNA. Within hours after injection of the DNA, IRBP protein is expressed in the liver.²⁵ Alternatively, we have transduced autologous B lymphocytes *ex vivo* with an IRBP construct using retrovirus, and infused them back into the mouse.²⁶ Both procedures resulted in a long-lived tolerance that prevented induction of EAU by a subsequent immunization with IRBP.

Thus, ocular immune privilege, which separates the healthy eye from the immune system, may, after a fashion, predispose to uveitis by impeding establishment of peripheral tolerance to retinal antigens.²⁷

The HOW

When a patient presents with uveitis, the physician has no way of knowing when and where the putative exposure to self antigen or a mimic antigen has occurred (with the exception of

sympathetic ophthalmia, a uveitic entity that follows a physical trauma to an eye and release of retinal antigens to the draining lymph node).³ However, we believe that we know the conditions under which exposure to retinal Ag or its mimic will result in disease. Immune stimuli from microorganisms and/or from damaged tissue signal “danger” to the immune system by activating pattern-recognition receptors on leukocytes and elicit an inflammatory environment. Antigen-specific immune responses induced in such an environment develop along pathways designed to destroy invading microbes. They also tend to be harmful to tissue and can result in induction of autoimmunity. In experimental animals, we purposely create such “danger” conditions by immunizing with retinal antigen emulsified in CFA, which contains heat-killed tuberculosis bacteria, so that it drives differentiation of retina-specific T cells to a pro-inflammatory phenotype.

Activation of retina-specific T cells occurs in the periphery, away from the eye, and they must reach and enter the eye for disease to be induced. This process is frequently referred to as “homing,” implying specific attraction to the target organ. Our data indicate that this term is a misnomer and that entry of the first activated T cells into the eye occurs at random. In experiments in which activated and fluorescently labeled retina-specific or nonspecific T cells were infused into recipient animals, a small but equal number of cells entered the retina within hours of infusion.^{27,28} While only the specific cells resulted in EAU development several days later, indicating that

induction of disease requires antigen recognition within the eye, this experiment shows that the first antigen-specific T cells entering the eye have no advantage over nonspecific T cells.

Since the healthy eye is protected by a blood-retinal barrier, which prevents free trafficking of cells and even of larger molecules, how are these first T cells able to enter the eye? It is important to keep in mind that activated T cells are sticky and invasive, somewhat resembling metastatic tumor cells. They express adhesion molecules, produce matrix-degrading enzymes, adhere to blood vessels and extravasate into tissues. Therefore, the blood-retinal barrier, which can stop quiescent naïve T cells from entering the eye, is ineffective against activated T cells. It is also important to emphasize that the retina-specific T cells that drive disease are by themselves not sufficient to inflict tissue damage. Recognition of their specific antigen within the eye maintains the activated state of the retina-specific T cells. The products that they secrete, including cytokines and chemokines, cause inflammatory changes in the retina, stimulate nearby ocular resident cells, and activate the retinal microvasculature. This attracts leukocytes from the circulation, which enter the eye in increasing numbers resulting in an inflammatory cascade that builds on itself. In this sense, the antigen-specific T cells are the orchestrators of the inflammatory process but it is the infiltrating blood-borne leukocytes/monocytes, granulocytes, natural killer (NK) cells, and NK T cells (NKT), as well as $\gamma\delta$ T cells, that amplify the inflammation and that mediate the final tissue damage.

Indeed, treatments that inhibit recruitment of blood-borne leukocytes into the eye (e.g., blockade of adhesion molecules, depletion of particular leukocyte types), also inhibit EAU. However, while treatments targeting inflammatory processes in general may be effective in ameliorating disease, they do not cure the underlying problem of breakdown in tolerance and presence of autopathogenic T cells, which will cause the disease to rebound as soon as treatment is discontinued. It is therefore of utmost importance to understand the nature of the eliciting retina-specific T cells to develop therapeutic regimens that target them effectively.

Current immunologic knowledge indicates that Ag-specific effector T lymphocytes fall into several major lineages that differ in phenotype and function, known as Th1, Th2, and Th17. They arise from a common precursor after different conditions of stimulation, and each produces a distinct profile of cytokines and chemokines, which is in line with their biological roles in dealing with different types of infections (Fig. 3). The natural role of Th2 cells (IL-4-, IL-5-, and IL-13-producing) is to deal with worm infections, Th1 (IFN- γ -producing) control intracellular microorganisms such as tuberculosis and Th17 (IL-17-producing) are needed for defense against extracellular microorganisms, such as extracellular bacteria and fungi.^{29,30} Importantly, each one of these cell types can also be involved in tissue pathology. Th2 cells are involved in allergy and asthma, whereas Th1 and Th17 cells participate in various inflammatory and autoimmune diseases such as uveitis, multiple sclerosis, rheumatoid arthritis, and psoriasis^{29,30} (Fig. 3).

Autoimmune uveitis in humans is a heterogeneous group of diseases that can differ in their clinical presentation and course, even though patients may respond to the same retinal antigen(s).³ Associations have been described with Th1 or with Th17 cytokines (reviewed in Refs. 29, 31) and may underlie heterogeneity. In human disease, the causal relationships are hard or impossible to prove, but results obtained in animal models support the notion that diverse cytokine response profiles may lead to ocular disease. This is exemplified by two distinct clinical and immunologic forms of murine EAU: the classic EAU model induced by immunization with IRBP (or its

FIGURE 3. Major T cell lineages in the CD4⁺ subset. The origin, characteristics, and function of the major T-effector and Treg cell lineages according to current knowledge. Data summarized broadly from existing literature. Highlighted in yellow are the signature cytokines of the subset.

peptide, IRBP₁₆₁₋₁₈₀) emulsified in CFA and a recently developed model induced by injection of IRBP₁₆₁₋₁₈₀ pulsed, in vitro-matured dendritic cells (DCs).^{19,32} Both forms of exposure to IRBP result in uveitis and retinal disease, but the two EAU models differ in fundus appearance, in clinical course and duration, in the nature of the inflammatory infiltrate recruited into the eye, which is monocytic in the classic CFA-EAU and granulocytic in DC-EAU, and most important, in their effector cytokine dependence: Th17 in CFA- and Th1 in DC-EAU. This last conclusion stems from experiments showing that treatment with neutralizing antibodies to IL-17, but not to IFN- γ , prevents and reverses CFA/EAU. Conversely, uveitogenic DC injected into IFN- γ ^{-/-} mice, which are unable to generate a fully functional Th1 effector response, fail to induce disease.^{19,32}

These experiments demonstrate that uveitis can be either Th17-driven or Th1-driven. The type of response that will predominate appears to be determined by conditions during first exposure to Ag, more specifically, by the quantity and/or quality of Toll-like receptor and other innate receptor signals as well as by the type and diversity of cells involved in antigen presentation (Fig. 4). Notably, Th17 becomes dominant when antigen is recognized in the context of CFA. We propose that the mycobacteria stimulate multiple microbial pattern recognition receptors on diverse cells residing in the lymph node draining the site of immunization. These are cells that participate in or influence the antigen presentation process and include DCs, monocytes, $\gamma\delta$ T cells, and possibly others. This intense and multifaceted stimulation leads to a Th17-dominant immune response. In contrast, Th1 becomes dominant when Ag is presented by in vitro-matured, antigen-pulsed DCs. Experimental evidence shows that in that situation the DCs themselves must migrate into the regional lymph nodes and present the antigen to the T cells (i.e., antigen introduced on the DCs is not transferred to host cells for cross-presentation).¹⁹ The DCs are matured before transfer, only with soluble endotoxin and anti-CD40 antibodies, resulting in a much more focused stimulus than CFA, leading to a Th1-dominant response. In

FIGURE 4. Conditions of initial exposure to antigen determine the nature of the subsequent T cell effector response in vivo. TLR, toll-like receptor; MTB, *Mycobacterium tuberculosis* bacteria; IRBP, interphoretic retinoid-binding protein; CFA, complete Freund's adjuvant; LTA, lipoteichoic acid; DC, dendritic cell; B, B cell; Mφ, monocyte/macrophage; LPS, lipopolysaccharide (bacterial endotoxin). Modified from Damsker JM, Hansen AM, Caspi RR. Th1 and Th17 cells: adversaries and collaborators. *Ann N Y Acad Sci* 1183:211–221.

human uveitis, these initial eliciting events are mostly unknown, as they occur long before the patient presents with disease. However, if our findings in animals are relevant to the human situation, these events could be critical to the course of the subsequent disease and would have direct implications for therapy. Thus, the Th1 pathway may be an appropriate target in some types of uveitis, whereas in other uveitic diseases, the focus should perhaps be on Th17.

The WHY NOT

If nontolerized, retina-specific cells persist in our bodies and can easily be activated, and given the apparent ineffectiveness of ocular immune privilege to protect against activated T cells, why does uveitis not occur more often? Experimental evidence suggests that this is at least in part due to presence of regulatory T (Treg) cells, whose role is to keep effector T cells (and perhaps not only T cells) in check. Treg cells broadly fall into two major categories. Natural Tregs (nTregs) which are generated in the thymus along with conventional T cells, by positive selection on their cognate antigen. Treg cells can also be induced (iTregs) from conventional T cells as part of normal regulation of all immune responses.

Treg cells involved in regulation of eye-relevant immune responses have been intensively studied in recent years. It has been known for a long time that ocular fluids can induce conversion of T cells to functional Tregs and that systemic induction of CD4 (afferent) and CD8 (efferent) Tregs occurs as part of the anterior chamber-associated immune deviation (ACAID) phenomenon.^{1,2,33,34} Given the apparent ability of the eye to induce its own regulatory circuits, it was not known as recently as 5 years ago whether the eye is dependent on other, more conventional, regulatory pathways. Our laboratory demonstrated for the first time that eye-relevant nTregs exist and determine the threshold of susceptibility to EAU.^{23,35} After depletion of nTregs by antibodies to the CD25 surface marker (IL-2 receptor), mice subsequently immunized for EAU develop markedly enhanced disease. We demonstrated that generation of IRBP-specific nTregs, unlike that of IRBP-specific effector T cells, requires endogenous IRBP. However, experiments using IRBP-deficient mice demonstrated that IRBP specificity is not required for nTregs to prevent uveitis. Paradoxically, the very mycobacteria in CFA that are necessary to support induction of EAU, also appear to directly or indirectly trigger activa-

FIGURE 5. Using the critical checkpoints in pathogenesis as targets for therapeutic interventions. Once identified, many of the critical checkpoints in pathogenesis of uveitis can be disrupted by specific manipulations. The interventions are designed to interrupt the disease process and/or augment the body's own regulatory mechanisms for prevention, disruption, or accelerated resolution of disease. Modified from Caspi RR. Ocular autoimmunity: the price of privilege? *Immunol Rev.* 213:23–35.

tion of Tregs that are not IRBP specific. Because only activation of Tregs requires a specific stimulus, once activated by mycobacterial components, Tregs of any specificity can inhibit generation of IRBP-specific effector T cells in the lymph nodes that drain the site of immunization.³⁵

Although specificity to IRBP may not be critical in the periphery if Tregs can be activated in another way, it is conceivable that IRBP-specific Tregs, natural and/or induced, would play a central role in the eye, where IRBP is present but mycobacteria are not. One way through which such Tregs could be induced is within the eye itself. Ocular fluids have long been known to convert conventional T cells to functional Tregs in culture, and recently such conversion has also been reported for T cells co-cultured with ocular pigmented epithelia (reviewed in Refs. 34, 36). Using IRBP TCR transgenic mice and IRBP TCR-specific peptide-MHC multimers, we are currently examining the ability of the living eye to convert conventional T cells to Tregs as well as characterizing the Tregs that infiltrate uveitic eyes of EAU-immunized mice for their antigen specificity, functionality, and role in natural resolution of EAU.

Importantly, T-reg cells can be induced by directed manipulations and can be shown to protect from EAU. An efficient way to induce Tregs is by hydrodynamic injection of naked DNA encoding a large fragment of IRBP, which is followed by expression of IRBP in the liver and induction of tolerance manifested as resistance to EAU induction.²⁵ It turns out that this tolerance is mediated at least in part by induction of Treg cells, which can be isolated from protected mice and expanded in culture by stimulation with immature DC+IRBP₁₆₁₋₁₈₀. When these expanded iTregs are infused into recipient mice that are subsequently immunized for EAU, they protect from induction of disease.²⁵ These findings suggest that therapeutic augmentation of T-regulatory cells could be explored as a future clinical approach.

It should be emphasized that Tregs are likely to account for only a part (albeit an important one) of the regulatory mechanisms that limit uveitis. Exhaustion of effector cells, induction of other regulatory cell types, including myeloid-derived suppressor cells (MDSC) and NKT cells. Induction of various negative feedback loops is also likely to play a role. One example, of non-Treg-driven regulatory mechanisms in which NKT cells may be involved is the apparent paradoxical role of IFN- γ in EAU. Although the (IFN- γ -dominated) Th1 response is clearly pathogenic and IFN- γ acting locally within the eye is highly proinflammatory,^{32,37} IFN- γ produced at the systemic level, especially at the time of disease induction, is protective in the model of IRBP/CFA-induced EAU. Thus, elicitation of high levels of IFN- γ by injection of IL-12 during the first week after uveitogenic immunization, but not later, aborts induction of EAU and inhibits its associated immunologic responses. Conversely, systemic IFN- γ neutralization enhances disease.^{38,39} Early induction of IFN- γ results in inhibition of effector Th1 and Th17 generation, possibly by their elimination through NO-dependent apoptosis.^{38,40} We have identified NKT cells as one of the IFN- γ sources whose early activation can control EAU.⁴⁰ This underscores the complexity of the pathogenesis of uveitis, in which a proinflammatory cytokine can also be protective, depending on the time, place, and target cells on which it acts.

IN SUMMARY

The basic mechanisms and critical checkpoints in the pathogenesis of uveitis that we and our colleagues have defined using animal models of the disease can serve as starting points to devise therapeutic strategies to interrupt the pathogenic

process (Fig. 5). Some therapies that are listed herein are already in use, whereas others are in various stages of clinical development.

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