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REVIEW ARTICLE

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Treatment of autoimmune diseases by inhibition of T-cell costimulation

Abstract Advances in our understanding of the mechanisms involved in immune activation and immune tolerance have laid the foundation for the development of new strategies for treating autoimmune diseases. In particular, the dissection of the two-signal process of T-cell activation has identified distinct targets that may provide a means of blocking pathological autoimmune responses without causing sustained blockade of protective immune responses. These strategies have shown great promise in animal models for autoimmune diseases, and they are currently the focus of clinical investigation in several autoimmune diseases of humans.

Key words Autoimmunity · Costimulation · CTLA4Ig · Murine lupus · Systemic lupus erythematosus

Introduction

Recent discoveries regarding the molecular mechanisms underlying T-cell activation and immune tolerance have paved the way for the development of new strategies to treat autoimmune diseases. Several of these strategies are based on the recognition that two signals are required for T-cell activation (Fig. 1) and that interruption of one of these signals may render selected T cells unresponsive.[1,2] The first signal occurs when T-cell receptors (TCR) bind to antigenic fragments on the surface of antigen-presenting cells (APC). This signal is required for T-cell responses, but it is not sufficient to activate T cells to mount an immune response. For that to occur, the T cell must also receive a second signal from other receptor–ligand pairs on the surface of T cells and APC. This second signal is referred to as costimulation, and its presence or absence determines whether a particular T cell will be activated to mount an immune response or, conversely, may be rendered unresponsive (anergic). This principle is at the heart of new therapeutic strategies that are designed to render autoreactive T cells unresponsive by blocking T-cell costimulation.

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The B7–CD28 pathway for T-cell costimulation

The ability to provide T-cell costimulation is not limited to a single receptor–ligand pair. Rather, several receptor–ligand pairs can contribute to T-cell activation. However, some of these interactions appear to be more critical to this process than others. In particular, the interaction between CD28 on T cells and the B7 family of molecules (B7-1 and B7-2) on APC plays a particularly important role, not only in the activation of protective T-cell responses but apparently also for the activation of pathological (autoreactive) T-cell responses.[3,4]

CD28 is expressed on virtually all T cells.[3] In contrast, the B7-1 and B7-2 ligands for CD28 are expressed in low density, if at all, on resting APC. They appear in high density on the surface of these cells only after the cells interact with Ag. As a consequence, it is anticipated that therapies directed against the B7 molecules would selectively affect cells that are in the process of Ag stimulation but would not affect resting cells. Thus, in people with autoimmune diseases, blockade of B7–CD28 interactions might preferentially inhibit lymphocytes that are in the process of responding to autoantigens without adversely affecting resting cells that are programmed to recognize other antigens.

To develop new agents that would block signaling via CD28, investigators have taken advantage of the homology between CD28 and another T-cell surface molecule, designated CTLA4 (Fig. 2). In contrast to CD28, CTLA4 is a negative regulator of T-cell function.[5,6] CTLA4 also differs from CD28 in that it binds to B7-1 and B7-2 with considerably higher avidity than does CD28.[7] Therefore, a fusion protein consisting of the extracellular domain of CTLA-4

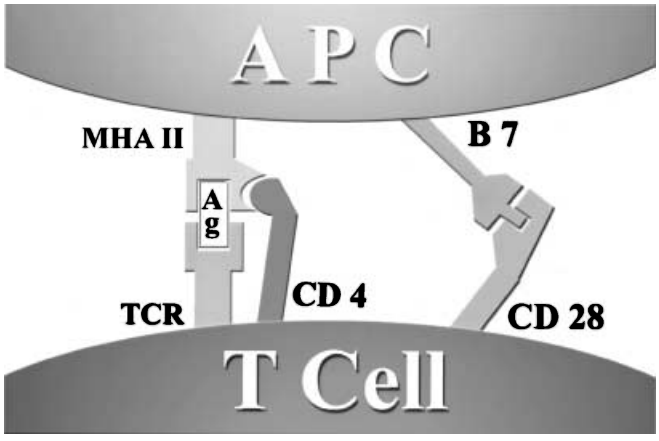

Fig. 1. Two signals govern the T-cell response to antigen. The first signal occurs when the T-cell receptor (*TCR*) binds an antigenic peptide (*Ag*) in the context of major histocompatibility complex (*MHA*) molecules on antigen-presenting cells (*APC*). The second signal involves other receptor–ligand pairs on the surface of T cells and APC, such as *CD28* on T cells and *B7* on APC

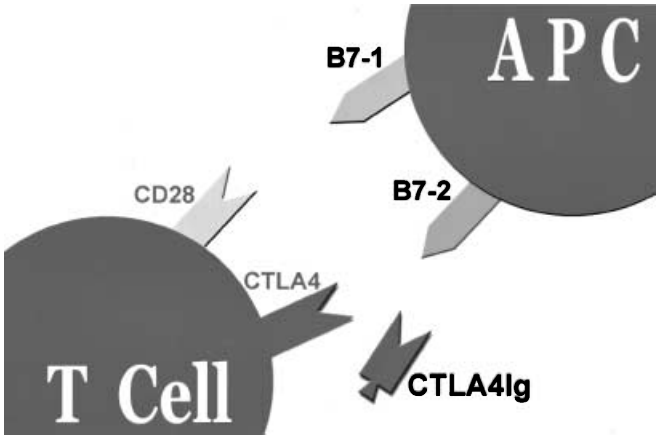

Fig. 2. One strategy designed to block T-cell costimulation is based on the homology between *CD28* and *CTLA4*. CTLA4 binds tightly to the B7 ligands for CD28. Therefore, a fusion protein composed of the extracellular domain of CTLA4 linked to the constant region of an immunoglobulin molecule (*CTLA4Ig*) can bind to *B7-1* and *B7-2* and, by so doing, prevent them from signaling T cells via CD28

linked to the constant region of an immunoglobulin molecule blocks the interaction between the B7 molecules and CD28 and thereby inhibits T-cell activation.[8,9] This fusion protein, designated CTLA4Ig, has been used successfully in mice to inhibit B-cell differentiation into immunoglobulin-secreting cells, to block T-cell responses, to facilitate organ transplantation, and to induce anergy to autoantigens.[10,11]

To test the potential value of CTLA4Ig in the treatment of autoimmune diseases, we treated lupus-prone NZB/NZW F_1 (B/W) mice with CTLA4Ig for 4 months beginning at the onset of disease.[12] Treatment prevented autoantibody production, reduced the severity of lupus nephritis, and prolonged life. Even when treatment was not begun until the late stages of disease, CTLA4Ig had substantial beneficial effects.[12] However, the benefits of treatment with CTLA4Ig were not permanent. Autoimmune disease was diminished for a period of time after cessation of therapy but then recurred.[13] Thus, although CTLA4Ig had considerable therapeutic value in B/W mice, it was not curative.

The CD40–CD40L pathway for T-cell costimulation

T-cell costimulation can also be provided by the interaction between CD40 on APC and CD40 ligand (CD40L) on T cells.[14] Like B7-1 and B7-2, CD40L is expressed on activated but not on resting cells. Thus, therapies directed against CD40L also have the potential to selectively inhibit active immune responses without having a generalized adverse impact on resting cells. The interaction between CD40 and CD40L contributes to T-cell costimulation in part by facilitating antigen-induced upregulation of B7-1 and B7-2.[14] It also stimulates proliferation, immunoglobulin production, and isotype switching by B cells.[14]

Similar to CTLA4Ig, monoclonal antibodies (mAb) that block costimulation via CD40L can suppress autoimmunity in murine models for several autoimmune diseases, including rheumatoid arthritis (RA)[15] and systemic lupus erythematosus (SLE).[16] For example, Mohan et al.[16] treated lupus-prone SWR/NZB (SNF_1) mice with three injections of mAb to CD40L at age 3 months, before the onset of autoimmune disease. Although treatment was brief, anti-CD40L delayed the development of lupus by several months. However, the mice eventually went on to develop lupus nephritis. Subsequent studies showed that chronic administration of anti-CD40L also had a beneficial, but impermanent, effect in B/W mice.[17]

Simultaneous blockade of the B7–CD28 and CD40–D40L pathways

The failure of either CTLA4Ig or anti-CD40Lto provide long-term inhibition of autoimmunity may reflect, at least in part, the fact that both pathways contribute to T-cell costimulation. Blockade of either pathway alone, therefore, may be inadequate to fully block costimulation. To test this hypothesis, we treated 5-month-old B/W mice for 2 weeks only with either CTLA4Ig alone, anti-CD40L alone, or both CTLA4Ig and anti-CD40L.[13] The benefits of treatment with CTLA4Ig alone or anti-CD40L alone were brief. However, in mice treated with both agents, the benefits of therapy lasted for a prolonged period without requiring ongoing immunosuppressive therapy (Fig. 3). As late as 10 months after cessation of therapy, 70% of the mice that received CTLA4Ig plus anti-CD40L were still alive, compared to only 18% survival among mice treated with anti-CD40L alone and 0% survival among mice treated with CTLA4Ig alone. These findings support the hope that brief blockade of T-cell costimulation, if sufficiently thorough, may provide long-lasting benefit in people with autoimmune diseases.

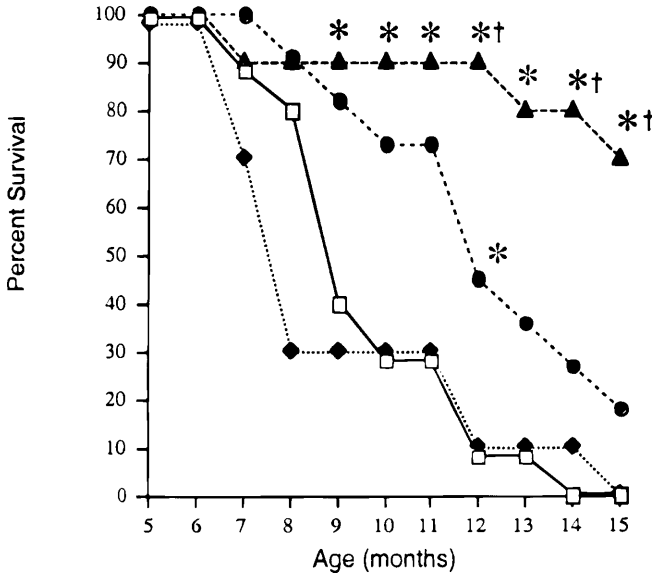

Fig. 3. Survival of female B/W mice that were treated for 2 weeks at age 5 months with either CTLA4Ig alone (*diamonds*), mAb to CD40L alone (*circles*), a combination of both CTLA4Ig and mAb to CD40L (*triangles*), or control Ig (*squares*). Asterisks (*) denote points at which there is a statistically significant difference from control mice ($P < 0.05$). The daggers (†) denote points at which there is a statistically significant difference from mice that received mAb to CD40L alone ($P < 0.05$). (Reprinted with permission from ref. 13, Copyright 1997, The American Association of Immunologists)

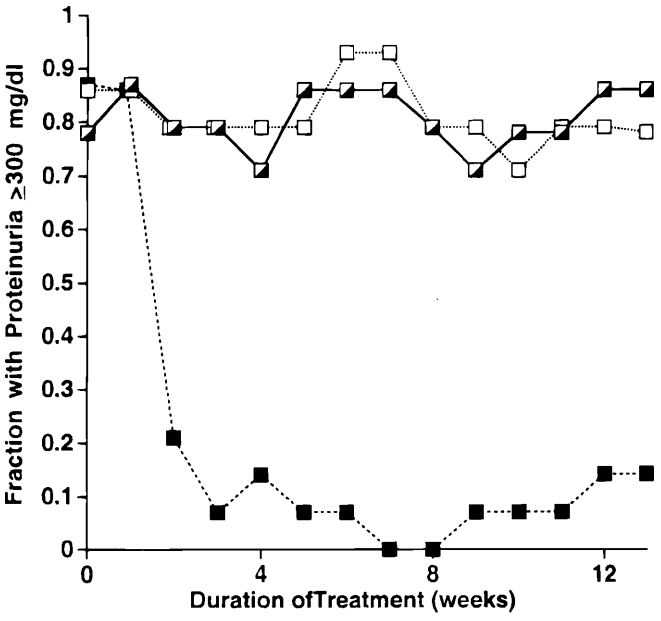

Fig. 4. Reversal of renal disease in female B/W mice with severe nephritis treated at age 6 months with either CTX alone (*half-solid boxes*), CTLA4Ig alone (*open boxes*), or combined CTX and CTLA4Ig (*solid boxes*). (Reprinted with permission from ref. 18, Copyright 2001, The American Association of Immunologists)

Simultaneous administration of CTLA4Ig and pulse cyclophosphamide

The combination of CTLA4Ig and anti-CD40L is not the only combination that has shown promise as a potential therapy for autoimmune diseases. CTLA4Ig and pulse cyclophosphamide (CTX) also act synergistically in the treatment of murine lupus.[18,19] Specifcially, we treated B/W mice with advanced renal disease with either pulse CTX, CTLA4Ig, both pulse CTX and CTLA4Ig, or saline.[18] After 12 weeks of treatment, the survival rate was 93% among mice treated with both agents, compared to 36% survival among mice treated with either CTX or CTLA4Ig alone, and 0% among control mice treated with saline (Fig. 4). Examination of renal pathology in a separate cohort of B/W mice treated in the same fashion established that treatment with both agents actually reduced renal injury from baseline, whereas treatment with either CTLA4Ig or CTX alone did not reverse renal injury but merely slowed the rate of progression relative to the saline-control mice.[19]

Blockade of T-cell costimulation in humans

Based on the encouraging results in murine models, two different mAb to CD40L have been examined in people with SLE. Initially, a phase I trial of one of the anti-CD40L mAb indicated that treatment was well tolerated, without any significant short-term toxicity.[20] This study employed a single-dose, dose-escalating design that was not intended to test efficacy. Subsequently, there have been two phase II trials of anti-CD40L in SLE, both of which have raised concerns about this approach. In one trial, 88 subjects with diverse manifestations of SLE were treated for 4 months.[21] Treatment was well tolerated, but it failed to produce statistically significant benefit in the primary clinical endpoint (reduction in SLEDAI score relative to control subjects). In a separate trial of a different anti-CD40 mAb, the occurrence of several unexplained thrombotic events prompted suspension of the trial before it could be completed. The mechanism responsible for the thrombotic complications has not yet been determined, nor is it clear whether this problem is limited to the particular mAb used in this study or whether it will apply generally to all anti-CD40 mAb independent of the target epitope. Studies are currently in progress to address these concerns.

The results of studies of CTLA4Ig in humans have been more encouraging. In a phase I trial of patients with psoriasis, CTLA4Ig not only was well tolerated but it also demonstrated clinically efficacy.[22] Based on the positive results in psoriasis, randomized, controlled phase II trials have been initiated in people with RA. If these studies succeed in reproducing the positive results that occurred in the psoriasis study, they will undoubtedly set the stage for more extensive examination of the effects of CTLA4g in other autoimmune diseases as well.

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