UPDATE

Releasing the Brakes on Antitumor Immune Response

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As is the case with firearms, the destructive capacity of the immune system must be carefully aimed and just as carefully regulated. A report by Leach *et al.* (*1*) in this issue is an encouraging example of how the receptors that regulate immune responses—in this case, CTLA-4, which was discussed in a recent Perspective (*2*)—can be directly targeted for immunotherapy.

T cell activation is normally self-limited so that the immune system returns to a baseline state once the offending antigen has been cleared. Initiation of T cell activation requires two signals, a major histocompatibility complex (MHC) antigen-specific signal delivered through the T cell receptor (*3*) and a "constimulatory" signal, the best characterized of which is delivered through the CD28 receptor (*4*). Each of these receptors has a counterregulatory cousin that recognizes the same ligand but delivers an opposing negative signal. The counterregulatory receptor for the T cell receptor is from the natural killer (NK) receptor gene family [first discovered on NK cells but also expressed on T cells (*5*)] CTLA-4 is the counterregulatory receptor for CD28; it binds B7 with roughly 10-fold higher affinity than does CD28. Occupancy of CTLA-4 directly counters the effects of CD28 on T cell activation (*6*): CD28 cross-linking increases lymphokine secretion by activated T cells, whereas CTLA-4 cross-linking decreases lymphokine production.

Full text of this and a previous Perspective (*2*) is available at http://science.mag.aaas.org/science/

Neither the NK receptor nor CTLA-4 is expressed on resting T cells; rather, they become induced after T cell activation. These delayed kinetics of expression are responsible for the characteristic self-limited nature of T cell activation. The importance of CTLA-4 in T cell homeostasis in vivo is most dramatically exemplified in CTLA-4 knockout mice. These mice develop a severe lymphoproliferative disease with immunologically mediated organ destruction (*7*).

Manipulation of these counterregulatory receptors in vivo has now entered the realm of cancer immunotherapy. Leach *et al.* have registered a success in this endeavor by injecting antibodies to CTLA-4 (anti–CTLA-4) into mice with tumors, thereby releasing the brake on T cell activation and favoring the accelerator. In two murine cancer models, infusions of antibodies to CTLA-4 produced an enhanced antitumor effect without overt toxicity. The success of this approach depends on the presence of a preexisting T cell response against the tumor antigens, which, under normal circumstances, fails to develop enough amplitude to win the battle against the tumor. This appears to be the case for at least one human tumor, melanoma, in which tumor-specific T cell responses can be readily identified in tumor-infiltrating lymphocytes (*8*).

CTLA treatment itself is not antigen-specific, and the window between development of an antitumor response and hyperimmunity or autoimmunity may be narrow. However, in contrast to genetic knockout, CTLA-4 blockade with antibodies allows a limited duration of therapy to take maximal advantage of even a small therapeutic window. Another intriguing possibility is the combination of cancer vaccines with short-term anti–CTLA-4 treatment. Cancer vaccines seek to present tumor antigens to the immune system in a fashion that favors T cell activation rather than tolerance. For a cancer vaccine to be successful, T cells specific for the vaccinating antigen must be activated above a threshold that provides them the edge over the tumor's growth. CTLA-4 blockade at the time of vaccination might allow selective amplification of immune responses against the vaccinating antigen while limiting induction of undesirable immune responses.

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References and Notes

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7. P. Waterhouse *et al.*, *Science* **270**, 985 (1995); E. A. Tivol *et al.*, *Immunity* **3**, 541 (1995).
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at the circled positions). Such a suppression result is very likely to mean that the splicing mechanism occurs by base pairing between U12 and the branch point region.

By inactivating U2, Tarn and Steitz (*4*) were able to obtain splicing of the AT-AC intron F from the human *P120* gene in nuclear splicing extracts derived from HeLa cells. With this in vitro system, they were able to map the branch point and confirm that it lies at the expected location within the TCCTTAAC consensus. Interaction between these nucleotides and U12 was demonstrated by psoralen cross-linking. These authors could also detect incorporation into spliceosomes not only of U12, but also U11 and U5. However, they were unable to detect U6, and inactivation of U6 did not interfere with P120 intron F splicing. This result is surprising in light of potential base pairing between U6 and U12 resembling that between U6 and U2 (*8*) and points to the existence of a minor snRNA related to U6. This unknown RNA should be capable of intimately associating with U12 and would also be expected to pair to the extended 5′ splice site consensus of AT-AC introns.

Like all results describing new phenomena, the two new reports raise more questions than they answer. Foremost among these is the extent to which the two mechanisms differ. We already know that U5 is present in both types of spliceosome and that both involve branch formation at a bulged A residue. Deeper kinship is suggested by an earlier result (*9*) that the yeast pre-mRNA splicing machinery (which is probably limited to the major class) shows some ability to recognize standard introns carrying mutations that alter the termini to AU and AC. Specifically, it had been known for some time that mutation of G at the first position of the intron blocked splicing after the first step. AC (and, to a lesser extent, A) at the last position of the intron can suppress this block (*9*). These and subsequent data [reviewed in (*10*)] argue persuasively for a non–Watson-Crick base pair between the nucleotides at the extreme ends of the intron that is important in the second step of splicing (see figure). These results imply that