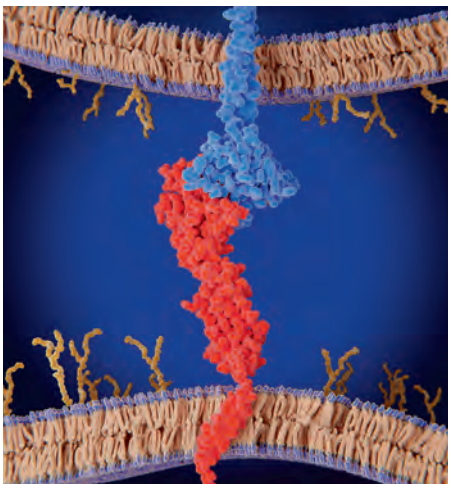
Milestone 13

Origin of immune checkpoint inhibitors

The 2018 Nobel Prize in Physiology or Medicine was jointly awarded to James P. Allison and Tasuku Honjo "for their discovery of cancer therapy by inhibition of negative immune regulation". Entangled in that description is far more history than the clinical development of immune checkpoint inhibitors for the treatment of cancer. First came the understanding of the fundamental nature and activation of T cells, principally to understand how tolerogenic mechanisms limit overactivity or self-reactivity, so-called inhibitory checkpoints.

The first characterized inhibitory immune checkpoint was the cytotoxic T-lymphocyte-associated antigen 4 (CTLA-4), so named by Brunet et al. at the Centre d'Immunologie INSERM-CNRS de Marseille-Luminy, France, where they discovered this immunoglobulin family member from a screen of cDNAs from mouse cytotoxic T cells. Subsequent work showed that CTLA-4, like the costimulatory molecule CD28 (Milestone 10), can bind B7-1 and B7-2 (known as CD80 and CD86 in humans). In 1992 at Bristol-Myers Squibb Pharmaceutical Research Institute, Seattle, Linsley et al. found that CTLA-4 is widely expressed not just by cytotoxic T cells but by activated T cells and is co-expressed with CD28. They also showed that anti-CTLA-4 antibody could block T cell proliferation in mixed leukocyte cultures, indicating that CTLA-4 might be another costimulatory molecule akin to CD28. As such, some confusion existed in the literature at the time as to whether CTLA-4 is naturally a costimulatory or inhibitory molecule. That is, until conclusive functional data came from a number of laboratories, first from the University of Chicago where Jeffrey Bluestone and colleagues demonstrated that blocking anti-CTLA-4 monovalent Fab fragments could augment T cell activation, which provided direct evidence that CTLA-4 is indeed a negative regulator of T cell function. At the University of California, Berkeley, Allison, then working with Max Krummel, further confirmed this conclusion in a 1995 paper. Meanwhile, Tak Mak and colleagues from the University of Toronto, as well as Jeffrey Bluestone working alongside Arlene Sharpe and colleagues at the Brigham and Women's Hospital and Harvard Medical School, Boston, discovered that CTLA-4-deficient mice develop T cell lymphoproliferative disease, which provided in vivo support for this mechanism.



"…conclusive functional data came from a number of laboratories, first from the University of Chicago…"

In 1996, Allison published the first in vivo evidence of efficacy for a checkpoint inhibitor as an anti-cancer therapy, showing that anti-CTLA-4 antibodies could clear carcinomas and fibrosarcomas from mice, which paved the way for these therapies to be used in humans and for his Nobel Prize.

The other half of that Prize came from a parallel line of study. Honjo and colleagues were studying programmed cell death at Kyoto University, Japan, and chanced on a protein they identified as highly expressed in the thymus. Expression of this protein was increased by anti-CD3 antibodies that the researchers were using to drive cell death in cell cultures, which resulted in the name 'programmed cell death protein 1' (PD-1). Later it would become evident that CD3-mediated T cell activation was the true signal that was driving PD-1 expression in these cultures, not cell death, but the name had stuck. But what was the ligand and function for PD-1?

In 1999, Lieping Chen's group at the Mayo Clinic in Rochester, Minnesota, discovered a third member of the B7 family that they named B7-H1. They showed B7-H1 is highly expressed in peripheral organs such as the lungs and thought this molecule was co-stimulatory. However, they also noted that it stimulates IL-10 production, which hinted at a regulatory function. At this time, they were unaware that B7-H1 is in fact the ligand for PD-1.

The following year, Honjo and the Gordon Freeman laboratory at Harvard plus others definitively identified the ligand for PD-1, without realizing it to be the same protein as Chen's B7-H1, and so named it programmed death-ligand 1 (PD-L1). They showed that PD-L1 is expressed by antigen presenting cells in non-lymphoid tissues such as the heart and lungs and provided the first clear evidence that this interaction can function as an inhibitory checkpoint.

Interestingly, those early parallel papers that showed differential anatomical expression of CTLA-4 (in lymphoid tissue) versus PD-1 (in non-lymphoid tissue) provided the first awareness that these two critical T cell inhibitory checkpoints function by distinct mechanisms. CTLA-4 is thought to function predominantly at the first point of antigen presentation for T cell priming, in order to limit overactive TCR–antigen–MHC responsiveness in the primary lymphoid organs. PD-1, by contrast, is more important in limiting tissue immunopathology by putting the brakes on effector T cells when they detect antigen in a target organ or tumour.

Together, these studies and many others not mentioned here have contributed to clinical trials and regulatory approvals for cancer, first with the anti-CTLA-4 monoclonal antibody ipilimumab (FDA approved for melanoma in 2011) and then with anti-PD-1 monoclonals nivolumab and pembrolizumab (FDA approved for melanoma in 2014). Today, although adverse events and low efficacy in tumours with low mutation rates are hurdles that must be overcome, many other checkpoint inhibitors have been approved for a wide variety of human cancers and are even in development for other pathologies such as autoimmune disease.

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Milestone study

Walunas T. L. et al. CTLA-4 can function as a negative regulator of T cell activation. *Immunity* **1**, 405–413 (1994)

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