Sensing self and non-self: New research into immune tolerance

February 13 2012, by Richard Harth

At the most basic level, the immune system must distinguish self from non-self, that is, it must discriminate between the molecular signatures of invading pathogens (non-self antigens) and cellular constituents that usually pose no risk to health (self-antigens).

The system is far from foolproof. Cancer cells can undergo unchecked proliferation, producing self-antigens that are tolerated by the immune system, rather than being targeted for destruction. At the opposite extreme, a range of so-called autoimmune disorders can result when healthy cells in the body are misidentified as hazards. The immune system has developed a further line of protection against such autoimmune responses in order to limit the pathology that can result. Essentially, the immune system is programmed to 'turn itself off' after prolonged recognition of an antigen.

In a new study appearing in the current issue of the journal *Science*, Dr. Joseph Blattman, a researcher at Arizona State University's Biodesign Institute examines how CD8 T cells—critical weapons in the body's defensive arsenal—are regulated when they transition from this tolerant state to an activated state and back. "We have previously shown that prolonged stimulation of T cells in disseminated cancers or chronic viral infections results in a tolerant state to the tumor or pathogen. It was never clear if this 'immune exhaustion' was a reversible fate or if 'resting' the T cells by removing them from the cancer or infection environment could restore their function" said Dr. Blattman. "These results show that even if you can temporarily rescue a tolerant T cell, it is hard-wired to

become tolerant again."

Lymphocytes or white blood cells are central players in the immune systems of all vertebrates, and come in various types. Large granular lymphocytes include natural killer cells (NK cells), while small lymphocytes consist of T cells and B cells. Cytotoxic T cells (also called CD8 T cells) take their name from their place of maturation in the thymus gland and the CD8 glycoprotein adorning their surfaces. These cells help the immune system identify infected or malignant cells and are the main cells responsible for eliminating them.

In the thymus, T cells undergo both positive and negative selection. In this process, T cells that bind too weakly or too strongly to self-antigens are weeded out, undergoing cell death. The first group would result in a deficient immune response to foreign invasion while the latter would tend to overreact to self proteins, leading to autoimmunity. Only about 2 percent of these developing T cells or thymocytes will survive. This dual process of selection generally produces cells capable of recognizing foreign threats while maintaining a tolerance for self-antigens.

However, some self-reactive CD8 T cells do make it out of the thymus and are exposed to self-antigen. In order to avoid causing autoimmune disease, the stimulation by self cells results in T cell tolerance. This could be for a number of reasons including lack of costimulation, the presence of regulatory T cells that inhibit CD8 T cell responses, or continuous stimulation by self antigens. This essential safeguard however can become an Achilles' heel, causing unresponsiveness in CD8 T cells to certain cancer antigens, many of which are self-antigens. One of the central challenges in tumor immunology is to somehow short-circuit T cell tolerance to tumor/self-antigens, without provoking autoimmunity.

Dr. Blattman and his group sought to illuminate the underlying molecular mechanisms of self-tolerance and the regulatory programs that maintain

or break it. Contrary to prevailing theory, the group demonstrated in a mouse model that T cells return to the tolerant state even in the absence of self-antigen. Further, such cells could be induced to proliferate and become functional if the lymphocyte cell numbers fell to appropriately low levels—a condition known as lymphopenia—and that this effect is observed even when self-antigen is present.

Because T cells are known to proliferate in lymphopenic environments, such as after chemotherapy and/or irradiation in cancer patients, the researchers used this to 'trick' T cells into proliferating in order to reset their function. This strategy did restore their function temporarily, but within a month afterwards, the T cells once again became tolerant even if they did not continue to encounter the tumor antigen.

The current research overturns a central paradigm regarding T-cell tolerance to self-antigens and may provoke a fundamental rethinking of the underlying mechanisms that govern the immune response. The results will help identify the molecular events that lead to T cell tolerance to tumor antigens, which should aid in development of strategies to permanently restore the function of T cells. This, in turn, should suggest new approaches for the treatment of cancer and chronic viral infections that employ adoptive transfer of modified cancer-specific T cells that make these cells resistant to becoming tolerant.

"Adoptive immunotherapy with T cells is an exciting strategy for combating cancer because the transferred T cells don't kill all dividing cells, but instead only target the cells expressing the cancer antigen. The problem has been that the transferred T cells usually become tolerant to the cancer" said Dr. Blattman. "By knowing the rules governing T cell tolerance, we will be able to identify what regulates this process and design ways of overcoming it in order to provide more effective cancer therapies."

The rescue of CT8 T cell functionality was indeed transient, in the mouse studies undertaken. When lymphocyte numbers in mice rebounded (having been reduced through irradiation), CD8 T cell tolerance snapped back into place, and the genetic master plan for these cells was reestablished. This fact implies that while a genetic blueprint oversees T cell tolerance, this characteristic is not entirely fixed, but may be subject to epigenetic regulation, that is, non-genetically encoded regulation that is transferred to each dividing cell.

Using techniques of genome-wide mRNA and microRNA profiling, Dr. Blattman and his colleagues uncovered a tolerance-specific gene profile for CD8 T cells, further demonstrating that this gene-based regulatory system could be overridden under lymphopenic conditions.

The rescue of CD8 T cells through this method has been dubbed homeostasis-driven proliferation. The mechanism operates even in the absence of a cognate antigen, but apparently shuts down once T cell homeostasis has been reestablished. Rescued T cells showed a reduction or down-regulation of tolerance-specific genes as well as an up-regulation of some 475 "rescue-associated" genes.

Evidentially, T cells are able to recall the tolerance program initially established after their first encounter with self-antigen, returning to it as a default, following repletion of lymphocyte numbers. While the precise mechanisms that account for this tolerant memory remain unclear, various forms of epigenetic gene regulation, not reliant on DNA sequence, are implied.

Future research will attempt to identify the signaling pathways associated with the interruption and reacquisition of T cell tolerance, which appears to operate independently of surface T cell receptors. Further, use of lymphopenia-mediated rescue of CD8 T cells for cancer therapy will require that tolerance-specific epigenetic memory somehow be erased.

Finally, lymphopenia-based T cell proliferation and activation also provides a model to describe heightened autoimmunity following organ transplantation, particularly cases of graft-host rejection. Such autoimmunity is often of a transient nature, again suggesting that T cell tolerance is reset once lymphocyte populations rebound following surgery.

"These results clearly suggest that epigenetic mechanisms are in place to maintain tolerance in T cells specific for self antigens. Uncovering precisely which key molecules and genes are important in this process should help us to improve T cell based approaches to the treatment of cancer, as well as to induce tolerance in T cells causing autoimmunity," said Dr. Blattman.

Provided by Arizona State University