

A Broader View of Immunotherapies

Fresh out of graduate school at Wayne State University in Michigan in 1985, Bernard Fox, Ph.D., landed a coveted fellowship with Steven Rosenberg, M.D., just as the first patients were being treated with cell-based immunotherapies at NCI. Now the Harder Family Chair for Cancer Research and Chief of the Laboratory of Molecular and Tumor Immunology in the Earle A. Chiles Research Institute at Providence Cancer Center, Fox combines basic and translational research to develop new forms of immunological interventions for cancer. Fox mentors students through the Oregon Health and Science University and has played a prominent leadership role in the Society for Immunotherapy of Cancer (SITC). He is also President and CEO of UbiVac, a company he cofounded to bring the tools forged from his research directly into the clinical armamentarium.

My laboratory is part of the Robert W. Franz Cancer Research Center, directed by Walter Urba, M.D., Ph.D. (who also spent 10 years at NCI), which has grown from three people, when we started 22 years ago, to 90 people focused on cancer immunotherapies. Currently, we are running 16 investigator-initiated trials, some of which are first-in-human studies. I've watched cancer immunology go from un-fundable to unstoppable, thanks in no small part to NCI.

My work is divided into a preclinical group studying basic mechanisms of T cell-mediated tumor elimination and developing therapeutic interventions in animal models, and a translational group that takes our discoveries into clinical trials and feeds back clinical information into the preclinical process. I manage a human applications lab that makes cell lines and products, which complies with practices required for administration to humans. The Center is also part of Bristol Myers Squibb's International Immuno-Oncology Network (II-ON), which brings together 10 leading institutions

to pursue clinical trials based on combination immunotherapies and multidimensional monitoring.

Bringing immunotherapies into the clinic is clearly not a solitary pursuit. It requires teamwork on multiple dimensions, across basic and clinical research, across nonprofit and commercial sectors, and across international boundaries. I view myself as kind of a bridge builder. When I was President of SITC, we brought together leaders of 15 different international societies to identify the major hurdles preventing the successful translation of immunotherapies. We identified issues ranging from limitations in animal models to limitations in training for scientists focused on translational research.

SITC has also been instrumental in supporting work across 13 countries to validate a prognostic biomarker in colon cancer that was first developed at INSERM. In 2006, Jerome Galon and colleagues published a remarkable correlation between the presence of specific T-cell infiltrates in an excised tumor and lack of recurrence. Such a biomarker, if validated, will not only have game-

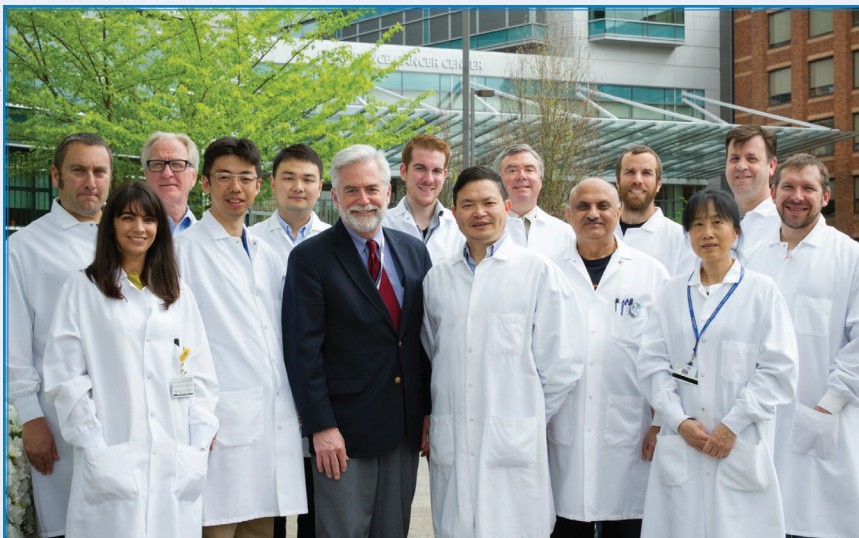
changing clinical implications, it also speaks to the increasingly clear link between the patients' immune response, therapeutically stimulated or not, and cancer morbidities.

Expanding Targets

We know that solid tumors are heterogeneous, continue to mutate, expand clonally, and spread to other parts of the body. As Bob Schreiber elegantly laid out in the Elimination-Equilibrium-Escape hypothesis, if your immune response is limited to a small number of targets, tumors will eventually escape from equilibrium. We probably aren't going to be able to find a single antigen to combat that diversity, but will need a broader immunotherapy strategy and multiple targets to which the host is not already tolerant.

In 2008, my former student and now colleague, Hong Ming Hu, Ph.D., developed a new vaccine strategy based on short-lived proteins (SLiPS) and defective ribosomal products (DRiPs). Normally, SLiPS and DRiPs are degraded by the proteasome and, we believe, are typically not cross-presented by antigen-presenting cells. When cells

(Photo: Courtesy of B. Fox)



Bernard Fox, Ph.D., and Hong Ming Ho, Ph.D. (center), and members of their laboratories at Robert W. Franz Cancer Research Center in the Earle A. Chiles Research Institute located within the Providence Cancer Center in Portland, Oregon

die and release proteins into the milieu, there are very few short-lived proteins around to generate peripheral tolerance. Hong Ming showed that if you blocked the proteasome (with bortezomib), SLiPS and DRiPs would be diverted into the autophagy pathway and end up in microvesicles which we know are studded with ligands for receptors found on antigen-presenting cells.

Moreover, we've shown that if you take these proteasome-blocked autophagic microvesicles from a tumor created in one mouse, you can use them to vaccinate another mouse against a somewhat related tumor. Back in 1957, Prehn and Main established that tumor-derived vaccines only protect against the specific tumor from which the vaccine was developed, and that has been dogma for 50 years. They took 10 identical mice (A, B, C, etc...) and gave them all methylcholanthrene to induce individual sarcomas. Vaccinating mice with tumor A only protected against tumor A, and the same was true of B, C, etc. In contrast, we were able to show that vaccinating mice with proteasome-blocked microvesicles from tumor A protects from a challenge with other sarcomas, in

eight of nine tumor combinations studied, changing the Prehn and Main paradigm. Much of this work was done in collaboration with Hong Ming's lab, and we continue to work together on several joint projects.

Our preclinical results led to support from NCI for a phase 1, and then a phase 2 clinical trial as an adjuvant study in definitively treated Stage IIIA/B non-small cell lung cancer, where the recurrence rate is 70 percent. We have reported on our first nine patients. All have developed strong immunity to many targets, which we know from The Cancer Genome Atlas (TCGA) are overexpressed in lung cancer. We have used mass spectrometry to study the protein content of the microvesicles and looked at whole exome sequencing. Our primary goal is to understand whether mutated or overexpressed nonmutated proteins induce the strongest antitumor responses.

With a new way to induce broad immunity, anti-PD1 or other costimulatory molecules could be synergistic. Some years ago, our institute developed anti-OX40, an antibody that can stimulate CD4 and CD8 T cells, as an anticancer therapy. We have shown in mouse models

that anti-OX40 boosts microvesicle-primed immunity in mice. We are very excited to move that data from the mouse into clinical trials.

The Patient Connection

There is a photo that I still see occasionally in papers, which shows the unbelievable shrinkage of metastatic melanoma nodules—it belongs to Linda Taylor (see "Immunotherapy's First Cure," *CCR connections* Vol. 8, No. 1). Taylor was treated with lymphokine-activated killer (LAK) cells and IL-2, shortly before I arrived at NCI as a Fellow in 1985, but she would come back periodically for monitoring. No one expected her remission to last 30 years. She is part of the linkage between laboratory and patients that was cemented for me during my years in Steven Rosenberg's laboratory. You knew that if it was promising, Steve would find a way to move your work into clinical trials. As a Ph.D. scientist, you can get busy and lose a little bit of that energy. So I try to make sure all my students have clinical experiences that relate to their translational projects.

Seven years ago, we had a patient with metastatic prostate cancer, they took him off ipilimumab when his enzyme levels spiked and he had a flare of hepatitis. However, he had a complete response and is still cancer free. Using protein arrays, we asked what proteins he was making antibodies against. One protein was the mitochondrial enzyme 3-hydroxyisobutyryl-CoA hydrolase (HIBCH) and we've identified HIBCH as overexpressed in his tumor. Less than 10 publications have referred to HIBCH, and none have identified HIBCH as a potential tumor antigen. The individual patients also drive us to think outside the box. With more time and resources to study these super-responders, eventually we will make everyone an exceptional responder.