QnAs with Bruce Beutler

Most people can tell when their bodies are in the throes of an infection, but Bruce Beutler is on a mission to determine where and how this recognition actually begins, at the level of microbe-sensing immune cell molecules. A professor of immunology at the University of Texas Southwestern and a recently elected member of the National Academy of Sciences, Beutler shared the 2011 Nobel Prize in Physiology or Medicine for demonstrating an infection-sensing role for the mammalian immune cell-surface receptor, TLR4. This protein activates the innate immune response—the body's primary line of defense against pathogens—upon detecting LPS, a structural component of certain bacteria. Beutler, who has long used genetic methods to tease apart the molecular details of the immune system, discusses his findings with PNAS.

PNAS: Your Nobel Prize-winning work began decades ago when you isolated from mice a factor known as tumor necrosis factor (TNF), which is made by immune cells called macrophages. How did that discovery lead to your research on TLR4?

Beutler: It was just quite by chance that I went to a laboratory as a postdoctoral fellow where a factor hypothetically associated with wasting in chronic disease was being isolated. I isolated that factor, which I found to be identical to TNF. I found that TNF was very strongly induced by LPS. I then made it my major goal to understand how LPS was sensed by macrophages of the host, because nobody really knew how microbes in general were detected during the first minutes and hours after inoculation. I tried many different ways to identify the LPS receptor, first using biochemical approaches, then expression cDNA cloning. These methods didn't yield any progress, so in the early 1990s I turned to positional cloning to try to identify the mutations that prevented all responses to LPS in two strains of mice. It took about 5 years, but my team and I tracked them down and showed that those mice had defects in the TLR4 gene.

PNAS: You emphasize the use of classic genetics—why do you think this is an important approach?

Beutler: I think that the power of the genetic approach lies in the fact that it's not biased. One uses a random process to generate phenotype, and in our case, a chemical mutagen, although there are other ways to mutagenize. One looks then for exceptional animals in which the phenomenon of interest—in our laboratory,

Bruce Beutler.

immunity—is disturbed in one way or another. One can track down the defect and find what protein was damaged to cause the observed phenotype; and that way, the real power, as I say, is the lack of bias. If one goes the other way and says, "I think for this, that, and the other reason that this protein is involved in this process"—in other words, makes a hypothesis—then one tends to have a personal stake in that hypothesis. Oftentimes scientists don't so much test the hypothesis as try to prove it correct, and that's how most errors occur in biology, in my opinion.

PNAS: Much of your previous work examined innate immunity. However, in your Inaugural Article, you focus on the antibody response, which is part of adaptive immunity.

Beutler: The two systems are related in some ways. There has been a theme out there that you must have an innate immune response to trigger the adaptive immune response that follows. We thought it would be good to examine this model using an unbiased approach. So we simply immunized mice in a uniform way, using either a T-dependent antigen or a T-independent antigen, and then looked to see whether the mice make an antibody response or not. In this way we found a total of 26 transmissible mutations. We tracked down and positionally cloned 19 of them successfully, and they fell into 17 genes, four of which were new and interesting. In fact, we found, quite to our surprise, that none of them operate in the innate compartment. Not

one. One possible interpretation holds that perhaps there's more autonomy to the adaptive immune system than people had realized. Alternatively, there might be a great deal of functional redundancy among genes required for the innate immune response.

PNAS: What avenues of immunological research does this open?

Beutler: We're very mechanistic in my group, and we tend to think of the immune system as a kind of a machine with cogs and wheels and gears. We see genetics as a way to get a complete list of parts, and to then begin to understand how they all fit together to give a humoral immune response, for example, or in the case of the innate immune system, to generate inflammation. We would like to find every component, and along the way, try to understand how the proteins fit together to do what they do.

There are of course several practical reasons to understand all this. One might be able to make better vaccines, for example; and there's a certain strain of thought that if only we had a better way to encourage an adaptive response at the level of the innate immune system, then we'd be able to make vaccines against malaria and HIV and all of the things that presently we don't have good vaccines for. Based on our results, one might begin to wonder whether this part of the response is really essential to trigger the adaptive response at all, or whether there might be other points of attack much further downstream that would lead to more efficacious vaccines.

PNAS: What do you think are the new frontiers in immunology?

Beutler: To me the greatest frontier in immunology is in the area of autoimmunity. The fact is that there's so much pathology caused by inflammation—everything that ends in "itis" has something to do with inflammation—and we don't know what causes most of it. We do understand that infectious organisms cause severe inflammation, and that has to do with how our immune system evolved to limit the spread of infection. However, what about sterile inflammatory disease? There, hardly anything is known, and I think that is a great frontier. When you talk about a common disease like rheumatoid arthritis, we know ways to treat it symptomatically, by blocking TNF for example, but where does the TNF come from? What really lights the fire? Nobody knows, not at all.

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