Moving from Cancer Genetics to Therapy

Dennis Slamon's early interest in understanding genes that viruses co-opt to become powerful pro-liferative agents ultimately led him and his colleagues and collaborators to the development of tras-tuzumab (Herceptin) for the treatment of HER-2-positive breast cancer. His work set the stage for current "givens" like genetic stratification of patients for treatment and using antibodies as thera-peutics. *Cell* editor Lara Szewczak spoke with Dennis, the recipient of the 2019 Lasker-DeBakey Clinical Medical Research Award, to discuss the research leading to trastuzumab and his views on how current progress in treating cancers may impact the US health care system. Excerpts from this conversation are presented below, and the full conversation is available with the article online.

Dennis Slamon
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Lara Szewczak: Thank you very much for taking the time to talk to me today, and congratulations on the 2019 Lasker-DeBakey Award.

Dennis Slamon: Thank you.

LS: You're sharing this award not only for the invention of trastuzumab or Herceptin, the antibody itself, but also for its development as a widely used therapy. I am curious: as a physician scientist early in your career, what got you interested in the receptor? What kicked it all off?

DS: I think what kicked it all off really was born out of work I did when I was doing my graduate work on the Special Virus Cancer Program back at the NCI. Everyone [got] interested in these acutely transforming retroviruses when they were looking for viral etiologies of cancers. And as it turns out, they really never found very many. The reality was studying those viruses that were causing cancers in experimental models gave us the first entrée to the fact that the reason these were the most potent oncogenic agents we'd ever seen, more than radiation or chemical carcinogens, was because they had very simple genomes, and they were carrying genes that were responsible for acutely transforming events. Those genes are ones that now everybody knows as Abl, Myc, HER-2 [ErbB-2], HER-1 [human EGFR, ErbB-1], and Src. These are all genes that these

retroviruses had stolen from normal cells. That led to the logical question of why would normal cells be carrying cancer-causing genes. They're not retained or conserved in nature because of that; they're conserved because they regulate growth. When they come under control of these viruses, they regulate abnormal growth in the process we call cancer. I was always interested in oncology, but then there was this molecular aspect that sort of got things kicked off.

LS: One of your key discoveries was that HER-2 was amplified in patient samples from many women. Is that correct?

DS: What I got interested in doing, which hadn't been done previously, was instead of doing this [work] in cell lines, was to actually take human tumors that were being removed for therapeutic purposes and to extract the DNA, RNA, and protein from them and to ask what differentiates what we're finding regarding these genes in tumor tissue versus the surrounding normal tissue.

In 1984, we published the first paper that led to the HER-2 collaboration with Axel Ullrich. We just looked at a number of different oncogenes in tumor and normal tissue from the same patient and found that there was differential expression. In 1986, Axel Ullrich knew what we were doing, I knew what he was doing, and he came and gave a talk. He had been one of

the first people involved in cloning, sequencing the EGF receptor [HER-1], insulin receptor, insulin-like growth factor, growth hormone, all in his early days at Genentech. What he had found, when he came to give the talk, was a sibling of HER-1, which was HER-2. It looked very much like [HER-1], and had a lot of sequence homology, but it wasn't identical, and it looked like a separate member of this receptor family.

It wasn't that we focused on HER-2. He had six different growth factors and growth factors receptors he had cloned, and I said, "Can we get those probes from you to do the kind of analysis we were doing with tumor tissue?" He absolutely agreed, and that started the collaboration. We walked through a number of different tumor banks we had, collections of 20, 30 tumors from different histologies—lung cancer, colorectal cancer, lymphomas, pancreas cancer. It wasn't until we got to the breast cancer panel that we found a hit that was obvious. We laid out 30 or 35 different breast cancers in that initial study and saw that for about 25% of them, loading the same amount of DNA in each lane, there was a dramatic increase in signal intensity, indicative of an alteration known as gene amplification.

That was exciting, but it was kind of a dead end because none of those samples had clinical correlates. I let Axel know what we found, that this was an alteration in breast cancer, that we didn't see it in some of the other cancers, didn't see it in the normal tissue. It wasn't germline because the normal tissue didn't have it. But I said what we needed was tumor tissues that had long-term follow-up and to see if we (A) see the same thing, and (B) if we do, does it correlate with anything? So that's how I started the collaboration with Gary Clark and Bill McGuire at the University of Texas, San Antonio. They had been studying estrogen receptor in breast cancer and had a collection of breast cancers with long-term follow-up.

I called them, told them what we found, and asked if we could get samples from them that we would analyze at UCLA. [We would be] blinded to the follow-up and make the call as to what's amplified or not and then send that data back to them, where they would run the correlation with outcome. I got a call back from Gary Clark soon after we sent the results saying there was a dramatic correlation with outcome, in that those patients whose tumors contained it [the amplification] had a much shorter disease-free survival. That was really an exciting moment that told us there was smoke there. This was a clear prognostic factor that was correlating with outcome

LS: What triggered the interest in pursuing an antibody approach to targeting HER-2?

DS: Well, we are thrilled that we're getting the Lasker Award and getting credit for this, but the truth is all of us stand on the shoulders of others. It is absolutely correct that Jeff Drebin, working in Mark Greene's lab at the time, was doing a parallel set of studies in an entirely different model. There was this homolog of the HER-2 gene in the mouse (Neu). He was studying antibodies against receptors and found that antibodies against the Neu oncogene could inhibit the growth of those cells *in vitro* and could inhibit tumor formation *in vivo*. So that was really the first clue. It was pretty exciting, but it still didn't prove anything, but it certainly said there was a lot more

smoke than we thought initially when we found the initial prognosis correlation.

The question was would it do it in human tumors? We set out on a series of experiments, and we did these at UCLA but in collaboration with Mike Shepherd. Axel by then had left Genentech and gone to the Max Planck to become director there. Mike took over the program, and Mike was very instrumental in the collaboration. In fact, [Mike was] one of the unsung heroes in the HER-2 story because he kept the program alive when a lot of people in the company weren't enthusiastic about it because antibodies had failed in the past and the assumption was this wouldn't work. But the only thing that was available to study the protein at the time were antibodies. Brian Finley at Genentech made a whole series of antibodies. Beatrice Langton did the same thing at a company called Triton Biosciences, and it was also done at Amgen. We got antibodies from everybody—Genentech's, Amgen's, and Triton's—and started testing and found that on those HER-2-positive breast cancer cell lines that were natural overexpressors, we could suppress the growth of those cells *in vitro* and stop tumor formation *in vivo*. In the ones that did not have [the amplification], you could put as much of the antibody as you wanted onto the cells and it would have no effect on their growth. So, there was specificity. This was exciting circumstantial evidence, but the final thing, at least for us, was to see [whether this would] happen in human breast cancer cells. We took human breast cancer cells that did not have the alteration, engineered them to overexpress the receptor at levels that you saw on patient material, and so mimicked within the lab what was happening in patients.

Sure enough, when we did that, those human breast cancer cells, which would grow in 2D and 3D and then make tumors, grew much faster *in vitro*, made more colonies in 3D, were much more tumorigenic in mice, and finally were much more metastatic. The metastatic potential increased by about 220%. That told us it was fire in human breast cancer. Then we took the antibodies and tested the series, including the ones from Genentech, and found that in the nine antibodies, I think three or four—but certainly three—had activities that could inhibit growth. That told us that this might be a therapeutic antibody.

LS: Two of the legacies of that work were the idea that you could stratify patients based on the genetic signature of their tumors and that antibodies could be therapeutic modalities potentially with broad applications.

DS: Genentech had just come off two big oncology failures: interferon and TNF. Now it's not that those drugs didn't have activity other places, but they were supposed to be blockbusters and that never went anywhere. There was not a lot of enthusiasm for another oncology protein therapeutic. Other antibodies had been tried against other antigens. Not necessarily active receptors, but other "tumor specific antigens," and they never really worked. The data that I just told you about kept coming in both what Genentech was doing internally and what we were doing externally. We worked very closely sharing all of our data in real time with them and they with us. It was a two-way street; it was a great collaboration in terms of the scientists. We were all excited about what we were seeing, but when we went to the people who have to make the

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decisions about whether [to] develop something clinically, they all came up with pretty much the same thing. You know, "We can cure a lot of mice, but we don't do much in humans." And we kept saying, "Yeah, but this is an active receptor, and it's altered in these tumors, and it's tracking with outcome. And in the preclinical models, when you put in the antibody, you reverse it, [and] that's gotta be tested in the clinic. This is more than just a marker." Ultimately that happened.

LS: If you fast forward, do you think there are still hurdles to developing and implementing antibodies as therapeutics?

DS: The real excitement, now, appropriate excitement, is in immune checkpoint therapy. A lot of people are pushing that, and the enthusiasm for antibodies is not as high as it used to be. Do I think that there are still antibody targets that can be approached? Absolutely. I think we're leaving some pay dirt on the table here, and then ADC (antibody-drug conjugate) technology, which has had its challenges, has now improved. In the HER-2 area, that is evidenced by the new Daiichi Sankyo drug that is an anti-HER-2 drug that works incredibly well in patients, who have failed other HER-2 therapeutics, including trastuzumab and T-DM1. So, I think there's a lot of excitement still, and I think there are other targets that the antibody approach can work for. Is it going to be everything? Absolutely not, but I think there are subtypes where the right surface antigens, or ligands for activating receptors, can be inhibited by antibodies.

LS: To come back to the patient stratification piece that now has spawned what we would call personalized medicine. You sort of go from the sublime to the ridiculous when you try to treat [patients]. Every cancer is one cancer on one end of the spectrum, and on the other, every cancer is unique and there is no such thing as a treatment across a population. I wonder what you think of that tension.

DS: Neither of the two extremes in terms of approaches are correct. We've known for decades that the genome of malignant cells is unstable by its very nature. That instability of the genome was baked in, which means that as the cancer cells are replicating, you get more and more errors, and there's tons of noise. In addition to noise, there are driver alterations. The

challenge is separating driver alterations that you want to target versus passengers that are just noise that you can target until the cows come home, and it won't do anything to the tumor. I see patients coming in saying, "I need to have my tumor sequenced." Yes, we can sequence your entire tumor; there's a whole industry now, of several companies, that offers this and does it. But how many times, honestly, has that information guided my clinical judgment, beyond what I knew using the traditional approaches we have with the markers that we know about now—Ras mutation for whether an EGF receptor antibody would work, BRAF mutation for melanoma, HER-2 amplification for breast cancer or gastric cancer, or ER/PR positivity for breast cancer that use hormonal therapy? All the rest of the information we get on all these mutations hasn't added a whole lot yet. That doesn't mean it won't, but it's still a research tool. It's not a therapy decision-making tool, for the most part.

LS: And do you think that time frame is 3 years, 10 years, 20 years? If you were a betting man, where would you put your chips?

DS: I would say within the next 36 months. All of that noise is not going to be clarified, but within all that noise we're going to start to pick out additional drivers and additional cassettes of things. We'll move from looking for one mutation that defines how the tumor is going to behave to a cassette of critical mutations. PI3-kinase mutation, in concert with HER-2 amplification, is associated with, in some instances, resistance to HER-2-directed therapies, so there is interplay with these signaling systems. Understanding that's going to give us a lot more options in thinking about how to combine therapies non-empirically.

LS: In basic research you focus a lot on, "What's a potential new target? What's a potential new lead? What might be druggable"? But there's a second side when patients have actually been treated. What then? There's a lot of care and support that needs to go on for patients because many of them need long-term treatment. I'm curious where you see the research, the health care ecosystem, and those patients.

DS: The ultimate objective is to cure cancers in patients even when they have advanced disease. Now that's a hard objective to obtain in many malignancies though it is obtainable in some. So, the fallback position, which isn't necessarily bad, is to take this from a lethal disease and make it a chronic disease that is controlled. Much like one can control diabetes. That requires active therapy that is ongoing for a long period of time. That now enters that whole new issue, that you can watch on the debates at night, of the cost of healthcare and these new therapies, which can be incredibly high. They're effective, and the ones that add really meaningful clinical impact should be approved, but then there's a burden on the healthcare system—if you have third-party payers on the insurers, if you have patients on the patient, if you have government paying on governments. This is a challenge in the health system when we change these into chronic diseases that require therapy long term, with these very expensive therapeutics.

I think there's going to have to be a reckoning and a realization that the for-profit issues that drive industry are appropriate, within reason. But I think that there have to be

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some limits set with what the drug does, how big an impact [it has]. This is not foreign to a number of countries where they make decisions on reimbursement based on how big an impact the drug makes in patient lives. It can be approved because the clinicals were all positive, but then they asked the question, are we going to fund it and pay it out of our healthcare and government insurance? That is something that is probably going to arrive in this country at some point because of the fact that we have to have some level of control of what the prices are, especially for these diseases that are going to be treated chronically.

LS: There's a complex ecosystem of government funding, private funding, philanthropy, patient advocacy groups. Where do you see those kinds of discussions and then ultimately monies coming from? What is the role for each of those?

DS: Well, I don't think you'll find too many people who would tell you that what we have now is sustainable. It's not. You can't spend 20% of the gross national product on healthcare. If you do, you better have the best outcomes in the world, and we don't. There's got to be a reckoning, and there's got to be a reconciliation that's going to involve the public and private sectors and government, and then that's going to require something that's pretty rare these days, which is working together. The British do it in their system to some degree, the Canadians do, the Germans do, in terms of deciding when the government's paying, what they're going to pay for, and what the impact of the drug is, and how much that is worth to society. In this country, probably, there'll be a public insurance thing that all people may have access to and private insurance, so I suspect it'll end up with a two-tier system. The current system, where everyone tries to have employer insurance covering part or all their insurance and then having these exorbitant copays or huge deductibles, that's just not sustainable for the kinds of drugs we're talking about.

LS: When you think about yourself and your colleagues who are similarly straddling patient care and drug development, what do you think your role is in that upcoming conversation?

DS: We should be advocates. On the physician side, we should be advocates for the patients. It, quite frankly, breaks my heart that there are people in the world today, less than before, but still, women who have HER-2-positive breast cancer. I mean we're talking 20% of breast cancer, and if there's 1.5 million to 1.6 million new cases a year, you can do the math. We're talking about 300,000 new cases of

HER-2-positive breast cancer a year. We know that the use of trastuzumab improves their outcome by 50% in terms of long-term survival, which is a big impact—more than we saw with chemotherapy, or any other therapy we've had, until hormonal therapy in the hormone receptor-positive patients came in. There are patients who have the disease, have the alteration present, and they don't have access to the drug because they live in places where the patient has to pay for it or in a country where there aren't sufficient funds for the government to pay for the drug, and the cost of the drug is still very high.

LS: The two-tiered system in the States that you envision happening, do you think that would exacerbate that situation?

DS: I don't think it'll necessarily exacerbate it. I think what'll happen is it'll introduce competition in the insurance market, including coverage of pharmaceuticals. That, if it behaves like other sectors of the economy, will drive prices down. Because if you can go into the public [insurance option], the government can negotiate with the drug companies for prices, like they do everywhere else except here. Then I think it'll be a competitive thing where the company still should make a profit from their investment and research and development of a new effective drug but that that profit has rational limits.

SUPPLEMENTAL INFORMATION

Supplemental Information can be found online at https://doi.org/10.1016/j.cell.2019.08.029.