Industrial R&D glimpsed

A significant minority of the technologies published in *Nature Biotechnology*'s pages originate from R&D in established biotech firms. One good example is an antibody linker technology developed at Seattle Genetics (Seattle) by Peter Senter. Numerous attempts have been made over the years to develop a linker that is stable enough in the bloodstream to last until an antibody-drug conjugate (ADC) reaches the tumor site, yet labile enough to deliver its payload effectively. Despite these efforts, only a single ADC has been approved. Mylotarg, developed by Wyeth Ayerst (Madison, NJ, USA) and approved for CD33-positive acute myeloid leukemia in 2000, consists of an anti-CD33 antibody linked to the antitumor antibiotic calicheamicin.

The conjugate works in part because it attacks a blood-borne malignancy and the antibody can find its target quickly, before its systemic drug release leads to decomposition and toxicity. "It can saturate the tumor within 30 minutes, so the requirement for stability is not nearly as high as for tumors that are less accessible," says Senter, who is vice president of chemistry at the company.

In his team's 2003 *Nature Biotechnology* paper[1], Senter and his colleagues describe a novel peptide linker designed to be cleaved by the enzyme cathepsin B, which is present within cell lysosomes. Most traditional linkers, including Mylotarg, are based on acid-labile linkers that are designed to release drug within acidic vesicles or in the acidic environment of tumors. However, they are somewhat unstable even outside of tumors, causing release of the active drug and hurting the conjugate's specificity.

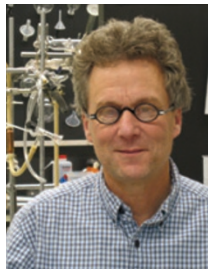
Seattle Genetic's ACD proved to be more specific and less toxic than previously reported ADCs, with a half-life in animals of 7–10 days, compared with typical half-lives of 1–3 days. Senter believes his company's approach addressed previous shortcomings. "A lot of the linkers used were too unstable. Others had flaws with the antibodies that were used. And a lot of the drugs used in the conjugates weren't potent enough to kill tumor cells effectively. We addressed each one of those problems in the 2003 *Nature Biotechnology* paper," Senter says.

The approach was taken forward by Seattle Genetics to develop a novel drug to pair with its linkers. It licensed a pentapeptide called auristatin E from Arizona State University natural product chemist George Pettit. Auristatins inhibit microtubial polymerization similar to how vinca alkaloids do, but more potently, Senter says. His team then developed synthetic derivatives of auristatin to prepare them for conjugation, and to tweak other properties.

"To go to a more stable linker required an understanding of the enzymes that are involved in drug activation, and precisely where they exist," says Senter. Biochemical studies showed that the primary enzyme responsible for cleaving peptide linkers is cathepsin B, which has minimal activity in the bloodstream. "That's why it's so much more stable than previous linkers." The combination of a stable linker and highly potent drug makes the conjugate both safe and effective, according to Senter.

The team compared the peptide linker–based conjugates to acid-labile conjugates. The peptide conjugate led to regressions and cures of tumors in mice at well-tolerated doses.

Although larger firms tend to regard promising technology as trade secrets, publishing Senter's work rather than keeping it under wraps was fully compatible with Seattle Genetics' strategy of raising company visibility, according to Peggy Pinkston, associate director of communications. To Pinkston the benefits are tangible. "Otherwise," she says, "companies [such as Seattle Genetics] couldn't attract ambitious and talented scientists."



Seattle Genetic's Peter Senter addressed shortcomings of antibody linkers, leading to a drug in clinical trials as well as revenue for his company.

The linker is a key component of one of Seattle Genetics' lead compounds, SGN-35, which is in phase 1 clinical trials for the treatment of Hodgkin's disease and other $CD30^+$ malignancies. The company is also collaborating with CuraGen (Branford, CT, USA), to develop a conjugate for the treatment of metastatic melanoma, which is in phase 1 trials. In total, the company has five other licensing deals centered around the technology, in which they provide the drug and the linker technology, and partners supply the antibody. Those licenses have brought in over $60 million to the company, accounting for essentially all of the company's revenue to date, according to Eric Dobmeier, chief business officer of Seattle Genetics. The company has also signed 50/50 codevelopment deals with Celera (Rockville, MD, USA) and Agensys (Santa Monica, CA, USA).

1. Doronina, S.O. *et al.* Development of potent monoclonal antibody-auristatin conjugates for cancer therapy. *Nat. Biotechnol.* **21**, 778–784 (2003). (57 citations)

Undersupported, but undaunted

Henry Daniell is not easily discouraged. After all, his invention—a universal chloroplast vector, which he first reported on in our pages in 1998 (ref. 1)—is now patented in 15 countries, thanks in no small measure, he thinks, to the validation it received by surviving the review process. What's not so good for Daniell and everyone else trying to commercialize a product from transgenic plants is the enormous cost of bringing such a product to market, especially considering the difficulty of obtaining the necessary capital.

But on the surface, things are going well. Recent work from his laboratory and elsewhere has demonstrated that human proteins like insulin and interferon can be made in plants with engineered chloroplasts at levels not possible with nuclear trangenes. In a later *Nature Biotechnology* paper[2], he reported that 45% of tobacco leaf protein was derived from a transgene, the insecticidal protein cry2Aa2, representing the highest level of foreign gene expression ever reported in plants at the time. Since then, equally impressive levels of insulin and interferon have been achieved using the system[3,4]. Low expression levels is the thing that Daniell feels has held back the field of plant-made pharmaceuticals—or at least one of the things. And with his chloroplast vector, which inserts into all 10,000 copies of the chloroplast genome, he feels he has that problem solved.

Another virtue of targeting the chloroplast genome with a transgene is that containment is not an issue. Because the chloroplast genome is maternally inherited, pollen won't disseminate the transgene. And with tobacco, one of the Daniell systems, at the time of harvest, there are no flowers (they harvest leaves), so no reproductive organs are involved, which should make regulators breathe a sigh of relief. In fact, four transgenic plants have been approved for field trials, and the results of one such trial (insulin) have recently been reported[5].

Daniell thinks he can further improve the odds of success with plants by delivering the expressed protein in plant powder. He recently showed that oral delivery of powdered plant cells expressing human proinsulin protected non-obese diabetic mice against development of insulitis. This could be a boon to