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T cells taken from a leukaemia patient and multiplied in culture are ready for infusion.

ADOPTIVE CELL THERAPY

Honing that killer instinct

Genetically altered immune cells are helping to push life-threatening cancers into remission and generating a buzz.

BY COURTNEY HUMPHRIES

A few years ago, when Michel Sadelain spoke about adoptive cell transfer (ACT) therapy at cancer meetings, his colleagues were dubious about what seemed a drastic and unconventional approach: harvesting and genetically altering his patient's immune cells to train them to attack her cancer. "I can't tell you how many nearly empty rooms I've spoken to about this technique," says Sadelain, director of the Center for Cell Engineering at Memorial Sloan-Kettering Cancer Center in New York.

The technique harnesses the power of the immune system by recruiting the body's own T cells — immune cells that recognize and marshal an attack against foreign invaders and diseased cells. T cells travel through the body, using their receptors to scan for small bits of protein called antigens on the surface of foreign cells. If an antigen matches the receptors, the T cell activates and launches an attack. In theory, malignant cells should be ideal targets for T cells, but tumours have ways of shielding themselves from an immune attack. With ACT, scientists tweak the T cells to give them a fighting chance. Sadelain calls them "living drugs".

Pilot studies in the past couple of years have had promising results, leading to increased interest and dozens more clinical trials investigating the technique. Success stories — albeit involving small numbers of patients — tell of people with aggressive cancers whose tumours melted away in days or weeks. In a field where extending life by a few weeks or months is considered a breakthrough, the complete remission of even a few patients is stunning. Sadelain is no longer speaking to empty rooms. Suddenly, he says, ACT has captured the imagination of scientists and pharmaceutical companies as if it were a new approach, rather than a field that has been developing for twenty years.

But there are both scientific and logistic challenges to expanding the use of this therapy. Researchers are still learning to control the cells' potency to ensure they can vanquish cancer without damaging normal tissue — an issue complicated by the fact that many cancer antigens are also found on normal cells. Another problem is that it's not yet clear how to turn ACT into a profitable business model, as harvesting and growing living cells requires much more time and skill than prescribing a drug. So while pharmaceutical companies are licensing proprietary receptors and looking into ways to scale up the process, that's just the start of the endeavour. As with any therapy, the companies still need to embark on large, multicentre clinical trials to test the effectiveness of the therapies on a broader group of patients. But large trials also require a way to engineer and distribute large quantities of cells, so they will only happen if companies are confident of long-term profitability.

Proponents of the approach say that the possibility of eradicating life-threatening tumours makes these challenges worth tackling. And recent progress in designing ACT therapies that are surprisingly effective is causing many in the field to sit up and take notice.

BOOSTING THE BODY'S CELLS

There are three strategies for ACT therapies (see 'Cellular attack'); the most-developed of which is the simplest. The tissue surrounding a tumour is likely to contain immune cells with antitumour activity, so doctors take a sample of this tissue and select those T cells that have been primed to attack the cancer. They culture these cells in the lab until they have enough, and re-infuse the cells back to patients along with the T-cell growth factor interleukin-2 (IL-2), which promotes the proliferation of antigen-specific T cells. However, the endogenous immune system has suppressive mechanisms that keep the immune response in check, and these mechanisms also prevent the newly transferred cells from working effectively. So patients must also be treated with drugs or radiation

to deplete their endogenous immune cells and allow the newly infused T cells to gain hold and fill the body.

This approach, called tumour-infiltrating lymphocyte (TIL) therapy, has been used successfully to treat only one type of cancer: metastatic melanoma. T cells that have been primed to attack a specific cancer are difficult to collect in a blood sample, but in melanoma these lymphocytes enter the tumour and are easy to biopsy. Over the past 25 years, a group led by Steven Rosenberg, an immunotherapy researcher and chief of surgery at the US National Cancer Institute in Bethesda, Maryland, has been building evidence that TIL therapy can alleviate or even eradicate melanoma in some patients.

"In the last of our trials, 40% of patients underwent complete, durable regressions of their melanoma," Rosenberg says of his latest results[1], published in 2011. Many of those patients had tumours throughout their bodies and had exhausted other treatments. This success vividly demonstrates how T cells sufficiently tuned to a specific cancer can have potent, long-lasting effects — and can even eradicate some tumours entirely.

But the current TIL regimen faces two big problems. First, patients must wait 4–6 weeks for the cells to grow before they can start therapy. The second problem is the need for specialized cell production facilities and staff trained in genetically modifying and growing the cells. Cassian Yee, a cancer immunologist at the University of Texas MD Anderson Cancer Center in Houston, says that work is being done to improve ACT and expand its use. New methods could make it possible to grow cells in days, rather than weeks, and research is underway to make it easier to obtain cells and to be more selective in the cells that are harvested. Yee and his colleagues have been developing methods to isolate tumour-specific cells circulating in the blood, for instance — a technique that could eventually make it feasible to treat cancers for which biopsies are hard to obtain or in which immune cells do not accumulate around the tumour.

KILLER CELLS

The success of TIL in melanoma is not currently transferable to other cancers, because it is harder to collect tumour-specific T cells. For those cancers, researchers are working to genetically modify T cells to hone their cancer killing skills. This strategy not only circumvents the need to find tumour-specific cells, but also allows scientists to tweak them in specific ways.

To do this, researchers are taking a couple of approaches. One option, called T-cell receptor (TCR) therapy, involves giving the cells new receptors that allow them to recognize specific cancer antigens; the receptors can even be modified to improve their ability to find and bind to their targets. To incorporate the new receptor, clinicians harvest a patient's T cells and then use a viral vector to deliver into the cells a gene that encodes the new receptor. The cells can also be engineered to express immune factors that prompt growth, that allow them to persist in the body, or that trigger other cells to attack the cancer. So far, TCR therapies have been shown to shrink tumours in some patients with metastatic melanoma, colorectal cancer and synovial sarcoma[2,3]. But there is one difficulty: the T-cell receptors must be genetically matched to the patient's immune type.

A more flexible tactic, called chimaeric antigen receptor (CAR) therapy, avoids this constraint. It uses a gene that encodes artificial, antibody-like proteins that bind the antigens studding the tumour cell's surface without needing to match the patient's immune type.

There are three pieces to CARs: an antibody that binds to a common cancer antigen; part of a receptor that activates the cell; and one or more stimulatory molecules that help the T cell proliferate and persist. When the CAR is inserted into and expressed in a T cell, it acts as a switch. As soon as the CAR encounters a matching antigen, it puts the T cells into attack mode. "The antibody provides the right conditions to find the tumour, and the T cell does the dirty work," explains Carl June, director of translational research at the University of Pennsylvania's Abramson Family Cancer Research Institute in Philadelphia.

Although conceived in the late 1980s, CAR therapies have only recently yielded positive results in small clinical trials. So far, they have all centred on the CD19 protein, which is expressed in B-cell leukaemias and lymphomas. CD19 is also expressed in normal B cells, which produce antibodies as part of the immune response. This means that CAR-initiated attacks can target healthy B cells, although the loss of these cells can be managed by therapies that treat antibody deficiencies.

In 2011, June and his colleagues reported that CAR T cells that target CD19 led to the remission of tumours in three patients with advanced chronic lymphocytic leukaemia (CLL) in whom multiple rounds of chemotherapy had failed[4]. Two of the patients experienced complete remission. In another study[5], led by Sadelain's group at the Memorial Sloan-Kettering Cancer Center, a different anti-CD19 CAR therapy led to remission for three of five adult patients with acute lymphoblastic leukaemia (ALL). Adult ALL is a terrible disease and the patients had already relapsed twice after chemotherapy, so these are "spectacular responses," Sadelain says. Researchers are now investigating whether CARs can be as effective against solid tumours as they are against blood cancers.

ON TARGET

Now that small clinical trials have shown that engineered T cells can effectively treat some forms of cancer, researchers must optimize the therapies to treat a variety of malignancies. Sadelain points out even these small studies show that different co-stimulatory molecules have different effects and, at least for CARs, some may work better for certain cancers than others. So part of the optimization process includes giving both TCR- and CAR-based T-cell therapies the optimal mix of enhancing molecules and targets to achieve the best response. Balancing components of the immune system to achieve the desired effect, Sadelain says, "is a completely new way of conceptualizing dosing in medicine."

Designing effective treatments requires finding cell-surface antigens for T cells to target without damaging normal tissue. This specificity may prove more difficult than researchers initially thought. Many antigens found in cancer are also expressed in normal tissue — HER2, for example, which is the target of the antibody-based therapy trastuzumab (Herceptin), is also expressed in heart cells. Before researchers can make progress, they must understand how extensively each candidate target is expressed in all tissues of the body.

Recent studies have highlighted what can happen when T cells unexpectedly attack normal tissue. In a clinical trial of TCR-engineered T cells, researchers at the US National Cancer Institute were targeting the cancer-specific antigen MAGE-A3 when two of their nine patients slipped into a coma and died. It turns out that the cells also recognized another member of the MAGE-A family that the researchers later discovered is expressed in low levels in brain tissue. Another type of

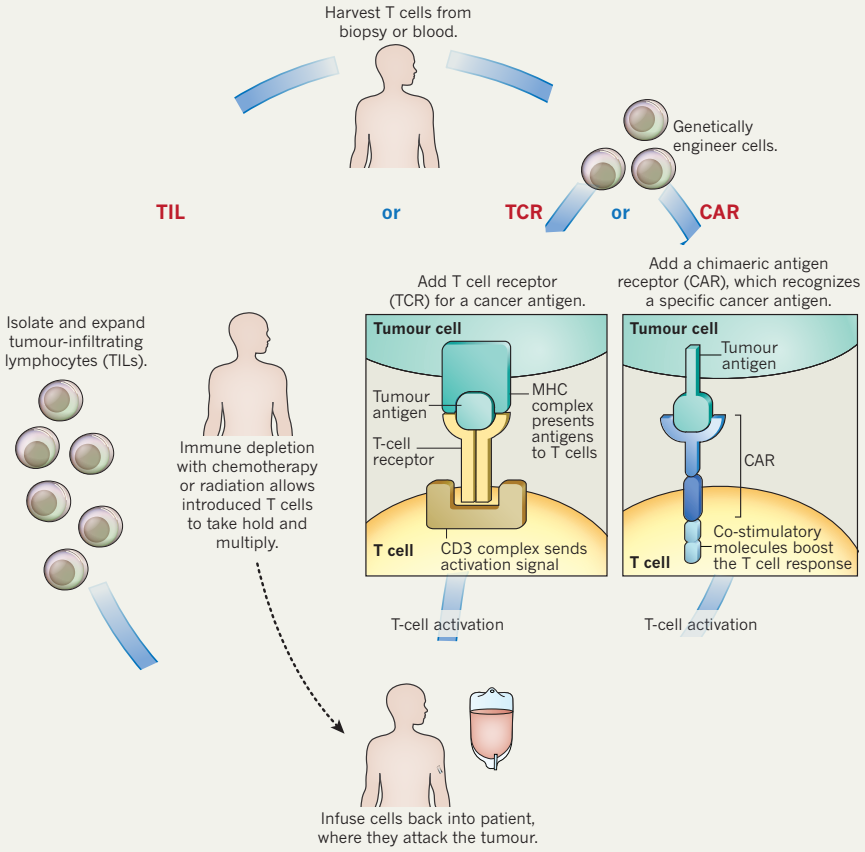
PUSHING AVAILABILITY

Adoptive cell transfer therapies for cancer are available only at a few medical centres right now, but several companies are working to change that.

Company	Therapy Type	Partnerships and trials
Adaptimmune	T-cell receptor (TCR)	Sponsoring nine pilot and phase I trials at several medical centres in the United States
Novartis	Chimaeric antigen receptor (CAR)	Licensing agreement with University of Pennsylvania to develop therapies. Establishing a global clinical trial program for CTL019, and expects to expand trials in the US next year.
Lion Biotechnologies	Tumour-infiltrating lymphocyte (TIL)	Licensing agreement with NCI to develop TIL for treating Stage IV metastatic melanoma
Kite Pharma	TCR, CAR	Agreement with the US National Cancer Institute to develop and commercialize products

CELLULAR ATTACK

Adoptive cell transfer (ACT) attacks cancer using either tumour-infiltrating lymphocytes (TILs) or genetically engineered T cells. Engineered cells are given either a new T-cell receptor (TCR) or an antibody-like molecule called a chimaeric antigen receptor (CAR); both activate the T cell when they encounter a particular cancer antigen.



MAGE-A3-specific TCR caused two patients to die from heart failure when the TCR bound to a similar protein, called Titin, which is expressed on heart cells. Adaptimmune, the company based near Oxford , UK, that developed the T-cell receptor, has implemented more extensive safety testing techniques in an attempt to prevent unexpected reactions in the future.

One of the main advantages of ACT is its speed — it works in days to weeks, much faster than other immune therapies — but triggering such a dramatic response can be dangerous. For instance, a patient with colorectal cancer who was infused with T cells as part of her CAR treatment died after experiencing an uncontrolled immune reaction called a cytokine storm. The process can also cause a condition called tumour lysis syndrome, which occurs when the chemical components of large numbers of dead tumour cells spill into the blood. "Our body is not built to get rid of 3–8 pounds of tumour," yet ACT therapies can do this in a matter of days, says Bruce Levine, director of the University of Pennsylvania's Clinical Cell and Vaccine Production Facility.

THE NEXT STEP

Both TCR and CAR therapies are being tested in patients with a variety of cancers, including ovarian cancer, pancreatic cancer, glioblastoma and mesothelioma, and results from these studies will help to determine whether the approach can be used more widely. Several unknowns remain, including why some patients get more therapeutic benefit from ACT than others. "Because you make the drug from a patient's own cells, there is variation at the source," Sadelain says. Some patients may have T cells that have lost potency or their ability to proliferate and that function more poorly. So studies must be done to find biomarkers that identify better-functioning cells, which could be used to predict patient outcomes, to sort cells before treatment, or to monitor treatment progress.

At the moment, ACT is a boutique therapy. It is performed in only a few academic medical centres worldwide, and has been tested mostly in small pilot trials in patients with advanced, chemotherapy-resistant disease. But it is progressing to larger trials. And because several groups have reproduced its success in the past few years, ACT is now drawing attention from the pharmaceutical industry (see 'Pushing availability'). But scaling up and commercializing a therapy that needs genetically modified cells will require cheaper, faster and more automated ways to modify and grow cells than currently exist. One company at the forefront of this work is Novartis, which has invested in such a facility to help it bring the manufacturing process used at the University of Pennsylvania to larger clinical trials.

Companies are largely focused on targeting common cancer antigens, such as CD19 and MAGE-A3, but not all researchers see this as the best approach. Rosenberg, for example, believes that the most successful strategy will be one that is totally personalized: engineering cells that target antigens unique to each patient's cancer and that are not found in healthy cells. Doing this would require extensive genetic analysis to find the tumour's unique mutations, he says, and then custom-crafting cells to match the cancer's genetic profile. Such an approach may be difficult, but he says that making an effective systemic cancer treatment is the priority. "Let's find out how to cure cancer even if it's very complex," he says, and then find a way to simplify it to treat large numbers of patients.

As ACT therapies move closer to the mainstream, the next big step will be investigating whether and how to integrate them with other cancer immunotherapies. In December 2012, the Cancer Research Institute, a non-profit organization based in New York that funds cancer immunology, joined forces with Stand Up To Cancer, a programme of the Entertainment Industry Foundation in Los Angeles, to award US$6 million over three years to a 'dream team' of researchers including Yee. The aim was to find out whether adoptive cell transfer can be effectively combined with another approach that's generating excitement in the cancer immunotherapy world: the use of immune checkpoint inhibitors. These proteins make the immune response to cancer more potent by removing signals — many of them released by tumours — that dampen the immune system (see 'Releasing the brakes', page S6). Pairing ACT with checkpoint inhibitors should simultaneously enhance the immune response and prime the immune system to attack the disease. "We've already had some preliminary data showing that the combination can be very effective," says Yee.

Despite lingering questions, Sadelain says that scientists and clinicians are enthusiastic about the potential of adoptive cell transfer. It represents a flexible platform for cancer treatment that can be tweaked and adapted as further discoveries are made. "This is not another small molecule or antibody," he says. "This is an entirely different approach to treating the patient." ■

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