

B-Cell Tumors Killed by T Cells Co-stimulated by CD80 and IL-15

Investigators at the Memorial Sloan-Kettering Cancer Center have engineered human T cells to recognize and kill cancer cells in a mouse model. In a paper published in the March 2003 issue of *Nature Medicine*, the workers transferred

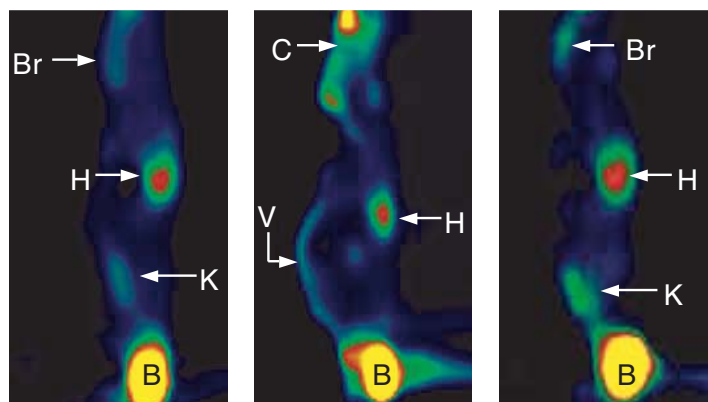
a gene encoding an antigen receptor into cultured human T cells, and then infused the modified cells into mice bearing tumor cells carrying the receptor on their surface. The

strategy was able to eradicate the artificial cancer in the mouse model. Furthermore, the researchers were able to show that transduced T cells from patients with advanced leukemia could effectively lyse autologous tumor cells *in vitro*.

The authors hope that the strategy will be tested in a clinical trial to treat patients with leukemias and lymphomas. "Our findings represent a step forward in the field of adoptive T-cell therapy," said senior author Michel Sadelain.

Earlier experiments had shown that genetically modified T cells could kill tumor cells *in vitro*, but the engineered cells were unable to proliferate and carry out their immunological mission once infused into mice. Crucial functions for this mission

include *in vivo* cytolytic activity and the ability to travel to tumor sites without prematurely succumbing to apoptosis. In the new study, the authors engineered T cells to target the CD19 antigen, a protein found on the surface of normal and cancerous B cells, such as those present in lymphoblastic leukemia, chronic lymphocytic leukemia, and most non-



PET scan of SCID-Beige mice. Left: tumor marker uptake in brain (Br) and heart (H), and excretion in kidney (K). Middle: mouse injected with tumor cells, showing tumor marking in the bone marrow (V) and calvarium (C). Right: mouse after T-cell therapy, showing loss of tumor marking, and kidney excretion of marker. Image reproduced with permission of Nature Publishing Group

Hodgkin's lymphomas.

Importantly, the engineered cells were expanded in the presence of CD80-expressing artificial antigen presenting cells and interleukin-15 (IL-15). T cells expanded in the presence of IL-15 persisted in tumor-bearing severe combined immunodeficiency (SCID)-Beige mice and eradicated disseminated intramedullary tumors. The anti-tumor activity of the cells was further enhanced by *in vivo* co-stimulation. "Collectively, these findings show that we have met many of the criteria necessary to conduct a clinical trial and test the approach in humans," said lead author Renier Brentjens, a member of Sadelain's laboratory.

Nature Medicine, Online edition, doi: 10.1038/nm827

Cardiac Myoblast Graft: A "Slow-Twitch Switch?"

New research has shown that myoblast grafts have survived and functioned following transplantation into a patient who previously suffered a myocardial infarction. In a paper published in the February 8, 2003 issue of *The Lancet*, Albert Hagege and colleagues injected cultured autologous myoblasts into the heart of a 72-year old man.

The aim of cell therapy after myocardial infarction is to regenerate cardiomyocytes from an exogenous supply of cells. Skeletal muscle myoblasts are a potential source of such cells. Transplantation of these cells directly into the heart has been proposed to improve heart function, but had not been directly demonstrated.

In the new study, the authors cultured myoblasts from the patient's thigh muscle and injected them into the unperfused scar tissue adjacent to the infarct. When the patient died from a stroke about 18 months later, the investigators were able to examine the injected area. They found well-developed myotubes with a conserved contractile apparatus. Interestingly, the myotubes expressed myosin isoforms associated with cardiac muscle despite their skeletal origin. This "slow-twitch switch" suggests that such cells could conceivably improve cardiac function.

The Lancet, 2003; 361:491-92. Doi not provided by publisher.

Technique Spins "Natural Bandage"

Investigators at Virginia Commonwealth University (VCU) have created a nanofiber mat that could eventually become a "natural bandage." Spun from strands of fibrinogen 1,000 times thinner than a human hair, the

fabric could be placed on a wound and never taken off. The research was reported in the Feb. 12, 2003 print edition of *Nano Letters*. The new material could be placed directly on the bleeding site to start the clotting process. Then, depending on the nature and severity of the wound, it could be left there to promote healing and eventually be absorbed by the body,

according to the researchers. It may be used for a minor cut or a battlefield wound, where it is vital to stop bleeding immediately.

“Or sometimes in surgery there are small bleeders that

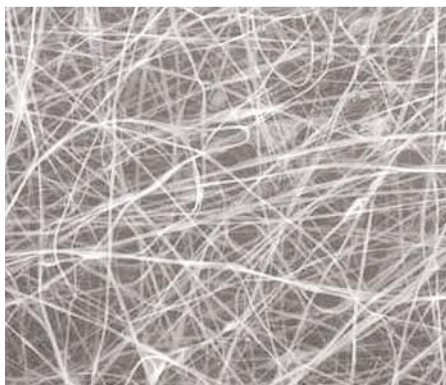
surgeons can't control,” says Gary Bowlin, lead author of the paper.

The mat is made from fibrinogen, a natural blood-clotting compound that is converted to fibrin by a cascade of reactions. “Fibrin is the meshwork, the netting,” Bowlin says. After the clot is formed and stabilized by the fibrin meshwork, that same meshwork sets the stage for the natural healing processes. To make the fibers, the researchers used a technique called electrospinning. The process begins with a solution of fibrinogen attached to a nozzle, which is then pointed at a metal target. An electric field is created between the nozzle and the target, and it is gradually increased until the force of the electric field overcomes the surface tension of the solution. This forms a liquid jet that is transformed into a dry fiber before it

reaches the target.

The solution is made with a high concentration that causes the polymer chains to intertwine. Instead of breaking into droplets just after the jet forms (which occurs in electrospray ionization), the jet continues as a continuous liquid stream. By the time it hits the target, the solvent has largely evaporated and fibers are formed.

“When the jet comes out, the polymer chains are tangled and help to form the fiber,” Bowlin says. “The key is that we're making these fibers at basically the same dimensions you would find in a natural clot,” Bowlin continues. VCU has licensed the technology to NanoMatrix, Inc.



SEM micrograph of the fibrous structure of the electrospun human fibrinogen scaffold composed of fibers 80 +/- 30 nm in diameter. Image courtesy Gary Bowlin.

Nano Letters, Online edition, doi: 10.1021/nl025866c

Homologous Recombination in Human ES Cells

A study in the March 2003 issue of *Nature Biotechnology*, by a team of scientists from the University of Wisconsin, reports methods for homologous recombination in human embryonic stem cells. “Homologous recombination is an essential technique for human ES cells to fulfil their promise as a basic research tool, and has important implications for ES cell-based transplantation and gene therapies,” write Thomas P. Zwaka and James A. Thomson, authors of the new study.

The technique has long been used

in the mouse. Significant differences between mouse and human embryonic stem cells have hampered the application of the technique to human ES cells, according to Zwaka. The research team was able to remove from the human genome the single gene that causes a rare genetic syndrome known as Lesch-Nyhan, characterized by an enzyme deficiency that manifests itself through self-mutilating behavior such as lip and finger biting and head banging.

The study of genes derived from human ES cells—as opposed to those found in mice—is important because, while there are many genetic similarities between mice and humans, they are not identical. There are human genes that differ in clinically significant ways from the corresponding mouse genes, said Zwaka. The gene that codes for Lesch-Nyhan is such a gene, as mice that do not have the enzyme do not exhibit the dramatic symptoms of the disease found in humans.

The new work may also speed the effort to produce cells that can be used therapeutically. Much of the promise of stem cells arises from their potential to differentiate into each type of cell in the human body. Marker genes can now be inserted into the DNA of human stem cells destined for a particular developmental fate, and the presence or absence of the gene should help clinicians sort cells for therapy. “Such ‘knock-ins’ will be useful to purify a specific ES-cell derived cell type from a mixed population,” Zwaka said. “It's all about cell lineages. You'll want dopamine neurons. You'll want heart cells. We think this technique will be important for getting us to that point.”

Nature Biotechnology, Online edition, doi: 10.1038/nbt788