PROMISE & PROGRESS

THE SIDNEY KIMMEL COMPREHENSIVE CANCER CENTER AT JOHNS HOPKINS

A Look Inside Our Cancer Medicine Cabinet

One-of-a-Kind Advances in Drug Discovery and Development

However, when it comes to limiting risk, the Kimmel Cancer Center's track record in drug discovery and development makes it a good bet. This success is attributable to its people, says Berger. "We have arguably the most knowledgeable researchers in the fields of cancer genetics, epigenetics and immunology, " he says. "Everything we enjoy today comes from basic science conducted 10, 20, 30, 40 or more years ago. Scientific discovery is not linear. The path forward is not always clear, but every finding adds to our knowledge and builds upon the foundation, and we have the experts and the willingness to collaborate that make it possible to put all of the pieces together."

It is this depth of expertise in all of the critical areas of cancer research and a culture that supports sharing information and working together that make it an incubator for new cancer drugs. The Kimmel Cancer Center is also a center that works lean and mean. "It is not the biggest cancer center, but it is, by any form of measurement, one of the most accomplished," says Berger.

The missing ingredient in what would otherwise be a nearly perfect recipe for drug discovery and development is sustained funding. Discovery is slowed because researchers get so far, but they have to stop and apply for more funding before they can move forward. Precious years are lost to the search for funding. "If we want to work quickly with focus, it takes funds that currently don't exist," says Liu. "If we had both, we could do more great things. "

Berger and Liu want the Chemical and Structural Biology Program to be that resource for cancer researchers. "We have to make drugs that attack one part of us without attacking another part," says Berger. "Investigators get stuck, and they don't know who to turn to."

The Pipeline

Immunotherapy discoveries are just part of the drug arsenal Kimmel Cancer Center scientists are helping to assemble. Nelson's approach is to go after every vulnerability of the cancer cell. He has been in the business long enough to know that the cancer cell is as complex and crafty as they come. With all of the natural processes of cell division and growth at its disposal, the cancer cell is a master at exploiting these processes to find new ways to cheat death. His vision is to use new technologies and reveal the genetic miscues that drive each person's cancer, and help find or develop drugs that either correct the miscues or shut them down.

There are always going to be a small number of cancers so dependent on a particular genetic miscue that they may only need one approach. This is exciting when it happens, but it doesn't apply to the majority of cancer patients. Most experts agree that cancers, particularly those diagnosed at an advanced stage that have had decades to corrupt many cell processes to their benefit, will likely require combined therapies, including surgery, targeted therapies, immunotherapies and radiation therapies.

"Gene mutations are like fingertips. You cut one off, and the cancer cells just work around it," says **Venu Raman**, who is working on a drug that attacks cancer cells directly and also sensitizes them to radiation.

Nelson believes a combined assault has the potential to disconnect cancer cells from their survival tools and finally overpower them.

RK-33

Raman's drug discovery began with research to understand the effect of secondhand smoke on breast cancer. It led him and his team to develop a first-in-class drug called RK-33. Countless hours in the lab and hundreds of experiments and assays later, Raman and his team have developed and patented a small molecule inhibitor of the DDX3 gene, an exciting first-in-class pharmaceutical.

Research that began in 2005 with funding from the Flight Attendant Medical Research Institute found that a gene called DDX3 was abundantly expressed in cells exposed to cigarette smoke. Raman's lab took a closer look at the gene, and when they blocked its function in animal models, tumors shrank, and the cancer didn't spread.

Tumors that spread from their original site, called metastatic, had the greatest expression of DDX3. "This finding fascinated me because metastatic cancers are the most difficult to treat," says Raman.

The DDX3 gene was already known to be instrumental in the replication of viruses, but no one had developed a way to block it. Working with a medicinal chemist, Raman came up with a series of potential drug compounds designed to inhibit DDX3 activity. After testing different combinations, the 33rd compound hit the target, and RK-33 was born.

"That was a big day," Raman says. "It's when everything went from theory to reality. We had discovered a new way to attack one of the key enablers of cancerous activity."

When Raman and his team first tested RK-33 in breast cancer cell lines, it had little effect on normal breast cells with low DDX3 expression. When they tested it in triple-negative breast cancer cells with high DDX3 expression, however, RK-33 easily killed the cancer cells.

"If you imagine your hand as a cancerous tumor," he says, referring to his mutation/fingertip analogy, "many of the drugs that we use to attack cancer act by cutting off a finger. There are still multiple other fingers left, and the rest of the hand can still function and evolve, leading to further spread and even adaptation of cancerous cells. RK-33 acts more like it is cutting off the wrist. When targeted successfully, it prevents a tumor's access to other survival options. Any tumor-sustaining mutations are rendered useless to the cancer cell because it cannot replicate itself."

Since RK-33 attacks overexpressed concentrations of DDX3 with greater intensity and efficacy, it should be toxic to tumors but not to the rest of the body.

VENU RAMAN IS WORKING ON A DRUG CALLED **RK-33** THAT ATTACKS CANCER CELLS DIRECTLY AND ALSO SENSITIZES THEM TO RADIATION. RAMAN'S DRUG DISCOVERY BEGAN WITH RESEARCH TO UNDERSTAND THE EFFECT OF SECONDHAND SMOKE ON BREAST CANCER. IT ALSO APPEARS TO BE ONE OF THE FEW DRUGS THAT WORK AGAINST METASTATIC CANCER CELLS.

Studies of the drug's toxic effects on normal cells followed, and as Raman increased the dose of RK-33, it began to work against cancer cells with different levels of DDX3 expression but did not harm normal cells. "We did extensive toxicology experiments," says Raman. "Even at four times the therapeutic dose, it was not toxic in animal models."

Since the project that originally led him to RK-33 involved smoking-associated cancer, Raman decided to also look at lung cancer, and he found it also had significant overexpression of DDX3.

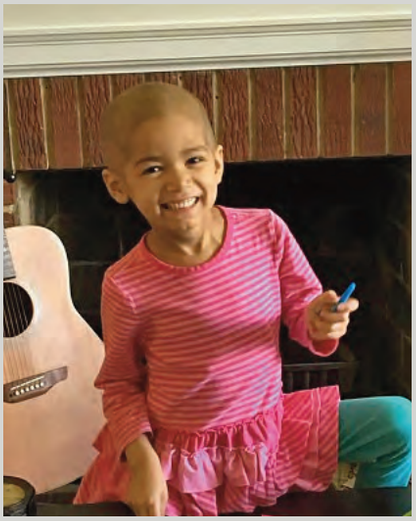
"I thought it was too good to be true," says Raman. "We repeated and repeated the lung cancer studies, and we found out that in sample after sample, this gene was overexpressed. It couldn't be a coincidence."

Further studies showed the gene was overexpressed in many cancer types, including triple-negative breast cancer, one of the most treatment-resistant forms of breast cancer; lung cancer; prostate cancer; sarcoma; and colorectal cancer. With his DDX3 gene target appearing to play a role across cancer types and his DDX3-blocking drug RK-33 patented and in development, Raman went back to the laboratory to decipher exactly how his drug worked at the cellular level.

"The gene is a critical part of the body's DNA repair mechanism," says Raman. "Cancer cells use it to reproduce and maintain the genetic stability essential to their survival." Raman found that blocking the gene with RK-33 not only killed cancer cells directly but also sensitized them to treatment with radiation therapy.

Radiation therapy kills cancer cells by damaging cell DNA beyond its ability to make repairs. Cancer cells that survive treatment do so because they are able to repair their DNA. Raman says RK-33 helps disable this repair mechanism. "If you irradiate cells, their DNA strands break, but over a short period of time, they get repaired. When you add RK-33, the strands remain broken. The cells cannot make repairs."

This finding led Raman to patent his drug as a radiation therapy sensitizer, but the evidence from his research shows it does more. One of the most exciting characteristics of RK-33 is its ability to destroy metastatic cancers—the often-lethal cancers that spread from the original site of a tumor and seed new, treatment-resistant tumors in different parts of the body. Metastatic breast cancers have very high levels of DDX3.



[Discovery]

Tara was diagnosed with advanced sarcoma two weeks before her 4th birthday. **Venu Raman** is working to get his experimental drug, RK-33, into clinical trials. In laboratory studies, the drug works well against metastatic cancer cells, and he is hopeful it will provide new options to patients with advanced cancers, like Tara. •

"Metastasis to the bone, brain and lung is common in cancer, but there are few drugs that have any long-lasting impact against metastatic cancers," says Raman. RK-33 could be the critical difference-maker in the fight against these entrenched, often terminal, cancers.

"Currently, there is no curative treatment for brain cancer metastasis," he says. "It's hard to find a silver bullet for cancer, but because RK-33 is nontoxic and a phenomenal radiosensitizer, there are so many opportunities, including metastatic cancers."

The potential to offer better outcomes to patients with the most difficult diagnoses is what Raman is most excited about. He offers a list of possibilities. "Advanced prostate and colon cancer, sarcoma (bone cancer), brain tumors, inflammatory breast cancer—all these indications are looking promising in multiple cancer models," he says. "We think RK-33 will work in any cancer that requires DDX3. And so far, all these difficult cancers require DDX3.

"My father is a colon cancer survivor," says Raman, "but unfortunately, not every patient responds to our current treatments. This compound represents a chance to change outcomes and save lives, and that's the best of what advanced biological research is about."

Raman is now in the last stages of refining RK-33. Because of its broad application, low toxicity and ability to sensitize cancer cells to radiation therapy, Raman wants a formulation that can be used in both adult and pediatric patients. His goal is to have a drug ready to go to patients in clinical trial within the year.

"We have a lot of pediatric cancer patients at Hopkins, and we are constantly looking to develop solutions that improve their outcomes," says Raman.

Raman draws inspiration from a particular patient. "Tara is a young girl who was diagnosed with a metastatic bone cancer called sarcoma two weeks before her 4th birthday," says Raman. "Currently, there is no standard of care for her disease because all treatments have worked so poorly. Nearly 80 percent of metastatic sarcoma patients relapse within two years of being diagnosed, and five-year survival is less than 20 percent.

"Hopefully this drug will offer new hope and better outcomes for patients like Tara and their families," Raman says. "That's why we're pushing to get RK-33 into human trials as fast as we can."

Despite these promising discoveries, Raman is now facing what is known as the "valley of death." Funding needs escalate rapidly as the drug is tested in humans and then across larger populations, and progress on the new drug will likely slow, or even stop, as he applies for more grants and appeals to more donors. His immediate goal is to obtain enough funding to complete the costly experiments required to file an Investigational New Drug application with the FDA.

The Flight Attendant Medical Research Institute, Safeway, the Dutch Cancer Foundation, Alex's Lemonade Stand Foundation, TEDCO and other funding partners have brought RK-33 this far, but Raman says that he needs about $3 million to $4 million more.

"Like it or not," Raman says, "the reality of the life sciences industry today is that the pace of getting new drugs to patients is controlled by investigators' ability to find financial partners."

Standing by anxiously are his Kimmel Cancer Center clinical collaborators: radiation oncologist **Phuoc Tran**, pediatric sarcoma expert **David Loeb** and breast cancer expert **Vered Stearns**, who will lead the clinical studies in patients.

"I am so lucky to work in a place like Hopkins. I have a great team working with me. From day one, all of them wanted to help—with no conditions. They are in it for the patients," says Raman. "That's essential because if you are trying to make advances against cancer, you need scientists and clinicians working together. I can't think of an institution that does it better than the Kimmel Cancer Center."

FLT3 Inhibitors

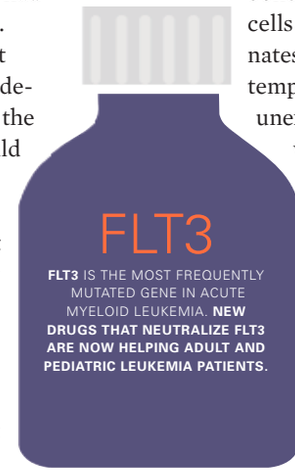
Pediatric Oncology Director and leukemia expert **Donald Small** understands the importance of bridging the laboratory and clinic to bring new drugs to patients. Working with **Mark Levis** in the laboratory and **Doug Smith** and **Patrick Brown** in the clinic, his FLT3 (pronounced flit three) discovery brought a new leukemia drug to adult and pediatric patients.

Small's research of hematopoiesis—how blood cells grow and expand—led him to clone the first human FLT3 gene. Next, he proved that it was very active in acute myeloid leukemia and some cases of acute lymphoid leukemia.

In fact, FLT3 turned out to be the most frequently mutated gene in acute myeloid leukemia. About one-third of patients diagnosed had the mutation—an alteration that made it almost impossible to cure them. "Having a FLT3 mutation reduces the chances of curing an AML patient from about 50 percent to less than 20 percent," says Small.

He had a target in FLT3, and if he could find a drug to neutralize it, Small believed combining such a drug with chemotherapy would improve cure rates for these patients, at least to rates of non-FLT3 AML and potentially even better.

Searching for such a drug proved to be a laborious, time-consuming process. He began by screening a library of more than 4,000 drugs known to target the family of proteins to which FLT3 belonged. He set up 96 wells, filled each one with FLT3-positive leukemia cells and tested each one of the 4,000 drugs to see if any of them killed the cancer cells. One by one, a specific amount of each drug was placed in the wells. "If the color changed, it meant the drug didn't work. If there was no color change, we knew we had an active drug," says Small. When he found a drug that worked, he systematically decreased the amount put in the wells to see how low he could take the drug and still get an anticancer response.



It wasn't high-tech, but at the time, it was the only way to get the job done, Small says. Now, there is an automated drug discovery tool called high-throughput screening that allows researchers to quickly perform millions of chemical, genetic or pharmacological tests, but in the late 1990s when Small began his research, low-tech was the only option for most university-based researchers.

When all of his tests were completed, CEP-701 stood out as the best drug. With a FLT3 inhibitor identified, Levis joined Small and began testing the drug in his laboratory and animal models to help figure out how to best use the drug in patients.

Since the drug had already been tested in clinical trials, this eliminated many of the FDA hurdles needed to move a drug into clinical trials, and Small and Levis partnered with Smith to take it to patients.

As Berger points out, drug discovery is not a single path. There is much back and forth between the laboratory and the clinic, following the science and the clinical data rather than a predetermined and straightforward path to get to the right drug. The essential ingredient of scientist/clinician collaboration is the reason the Kimmel Cancer Center is the perfect environment for drug discovery.

"We're not as big as other places, but we're really, *really* good at working together," says Smith. "We're also outstanding at basic science, clinical research and clinical practice, and that's the translational machinery that makes drug discovery and development possible."

When Smith took CEP-701 to patients, it was a mixed bag of results. The drug cleared leukemia cells out of the bloodstream and, in some patients, out of the bone marrow where new blood cells are made and leukemia originates. But, the responses were temporary, a result not completely unexpected in phase I trials where the sickest of the sick are typically treated.

Levis developed an assay for FLT3, a test that tells if the drug is actually hitting its intended target. "We were excited to see it was killing leukemia cells, and we had an assay to measure it, but we still needed to dig deeper," says Levis.

The group's goal was to get patients into remission using a combination of a FLT3 inhibitor and chemotherapy so they could receive a bone marrow transplant, a potentially curative therapy that replaces the patient's diseased bone marrow with healthy marrow from a donor. Levis' assay proved the drug was hitting its target, but in larger studies, it also showed it had a serious flaw.

The target was a good one, but in many patients, the drug's chemistry allowed the proteins in their bodies to suck up too much of the drug before it hit its target.

Levis went to the inpatient unit, watched patients take CEP-701, got blood samples from patients, carried the blood back to his laboratory himself and then used his assay to test the samples to see if the drug was hitting the FLT3