BORN IN T

by Kathleen Yount

photography by Jared Lazarus

Liquid tumors like leukemia, lymphoma, and myeloma are notoriously unpredictable—but genomic research is yielding intelligence that may corner these elusive cancers at last.

THE BLOOD

One of Duke oncologist Sandeep Dave's favorite stories is that of a professor at his medical school. "He was the kind of guy who could run up the stairs and leave all the young students huffing and puffing behind him," Dave says. Unbeknownst to most of these winded students, the physician was also suffering from follicular lymphoma, which he was monitoring without treatment, according to his oncologist's recommendations.

One day on rounds the doctor suffered a heart attack and underwent emergency bypass surgery. "The recovery was terrible," says Dave—infection set into his chest incision; he was "essentially at death's door" and remained hospitalized for three months. He eventually recovered and returned to work—and when he visited his oncologist a few months after the ordeal, bloodwork showed that his lymphoma had completely resolved.

Spontaneous remission can occur in a small minority of patients with follicular lymphoma—somehow the body's immune system wins the duel with the lymphoma cells that are attempting to overtake normal, healthy white blood cells in the lymph nodes and elsewhere. But a number of patients with the same disease will die within months of diagnosis. This sort of slippery prognosis makes the term *liquid tumors*—which includes leukemias, lymphomas, and multiple myeloma—especially apt. These hematologic cancers develop in the marrow of our bones, inexorably squeezing out the healthy cells in the blood that nourishes every tissue in our body. They can't be excised by surgery; there are no known effective screening methods or reliable ways to reduce one's risk. And the number of patients with these cancers is rising.

Duke hematologic clinicians have achieved many of the greatest successes in liquid tumor treatment, from improving bone marrow and cord blood transplantation to testing targeted drug therapies such as imatinib (Gleevec). Meanwhile, Duke's discovery of the breast cancer mutation on the BRCA-1 gene and the development of the Institute for Genome Sciences & Policy have put Duke at the forefront of genetic cancer research. Nationally respected hematologic clinicians like Joseph Moore, MD, have built a sizeable patient base, while hematologic researchers like J. Brice Weinberg, MD, have built vast stores of tumor samples for

analysis. "We've had excellent research and excellent clinical care," says Duke hematologic malignancy program director David Rizzieri, MD, "but we haven't always had a good bridge between the two."

Over the last 10 years, though, the program has worked to link its large patient population with its prolific bench research. Much of this translational bridge has been built over the ever-swelling current of genomic discovery: a broadening understanding of exactly what genes are aberrantly active, or overexpressed, during the genesis of cancer. This type of analysis paints a portrait of a tumor in detail previously unavailable to researchers; for each tumor, it unveils a palette of overactive cellular processes, or pathways, that essentially make that tumor tick. Dave says genomics makes possible the blueprint for personalized medicine: cancer treatment that begins with genomic profiling, so that patients receive only the therapies to which they are likely to respond. "It seems very commonsense," Dave says, "but it's far from the standard right now. The standard practice is that if the patient fails one treatment, he gets something else, then something else, then something else. And the results are highly variable—in many cases they are quite, quite poor."

Several years of genomic research have brought oncology to a watershed moment, Rizzieri says, with clinical trials of genomic-based therapies popping up in program after program. Now the youngest generation of liquid-tumor researchers at Duke carries the charge of walking this line between the bench and the bedside, to speed the translation of discovery into therapy.

Bettering the best

Chronic myelogenous leukemia, or CML, made headlines when Gleevec hit the streets in 2001. Hailed as a miracle worker, Gleevec (imatinib) has been quite effective in many patients by blocking a certain chemical pathway. But the drug is no panacea—it is not curative and a significant number of patients grow resistant to its effects. One of the drawbacks to Gleevec, says Duke cancer biologist Tannishtha Reya, PhD, is that even in patients who don't develop resistance to the drug, it doesn't affect the cancer stem cells, which are the cells that propagate the cancer. "So you always have to be on the drug," says Reya. Her team has been searching for a new chemical pathway that could be targeted to attack the cancer stem cells and sidestep the biologic roadblock that halts Gleevec's usefulness in some patients. And they've hit a potential treatment jackpot: a pathway called Hedgehog, which is known to be active in many solid tumors. "There are currently several drugs in development that can block this pathway in solid tumors," Reya says, "so it was a really unique opportunity to see if the approach would be effective in leukemia." First the team studied mice that were genetically altered to lack the Hedgehog pathway at birth. The mice had a significantly reduced incidence of CML, and those who did develop CML showed both delayed disease progression and a reduced number of cancer stem cells. Reya's team then tried blocking Hedgehog in normal mice using a drug called cyclopamine, a small molecule inhibitor that can be delivered easily into the body. Half of the mice treated with cyclopamine survived the cancer, while all of the mice that were not treated succumbed to the tumor. So Reya's group worked with David Rizzieri and John Chute, MD, to test the effects of cyclopamine on tumor samples from human patients who were in an advanced phase of CML. "The human samples have been remarkably responsive," she says, and the team is now planning to further test Hedgehog inhibitors; if the studies bear out, she says, they could open a new window to therapies not only for CML but also for other liquid tumors.

Examining the anomalies

The CML treatment successes of the last decade grow more impressive when compared to current therapies for other forms of leukemia, many of which have been idling essentially unchanged since the late 1960s. The standard therapy for patients with acute myelogenous leukemia, or AML, is what's called the 7 and 3 regimen: a strategic pairing of two powerful chemotherapies to create a treatment that is aggressive, toxic—and in many cases simply ineffective.

David Rizzieri, director of Duke's hematologic malignancies program, says genomic research has brought liquid tumor treatment to a watershed moment.

"We've been getting the same abysmal results for years," says oncologist Arati Rao, MD. Only one in five patients diagnosed with AML lives five years after diagnosis; most die within a year or two. The prognosis is even worse for older people, who make up the bulk of AML patients and who have a two-year disease-free survival rate of less than 10 percent.

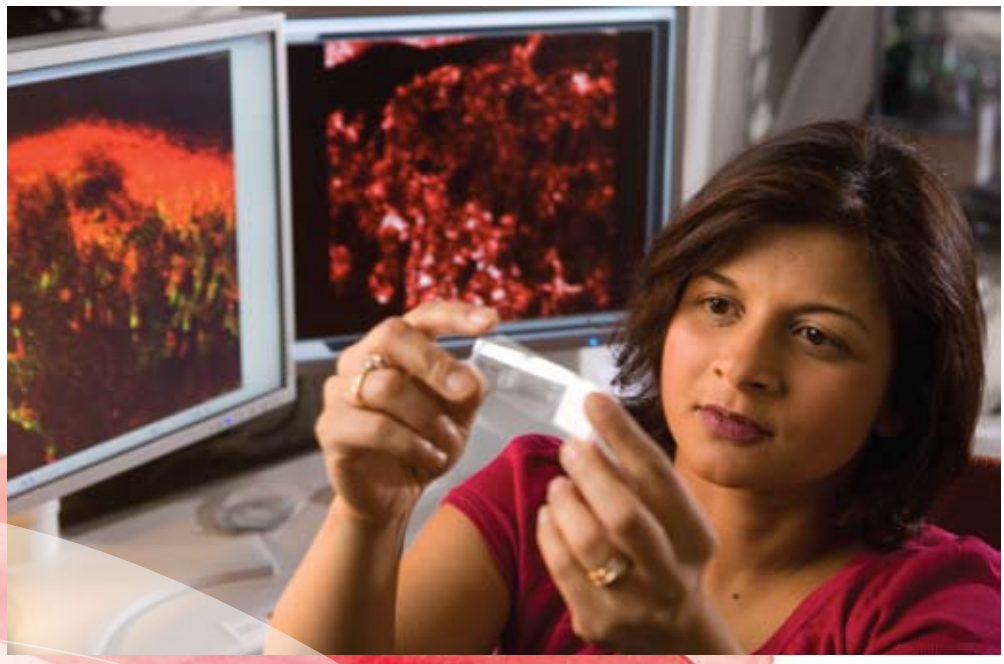
Rao is working to get to the biological bottom of what makes older AML patients so much tougher to treat. She and colleagues in the Netherlands and Germany gathered a cohort of 425 patients and studied patients who were 45 and younger and 55 and older. The clinical differences between the two groups were considerable: most patients in the younger group responded to therapy, and they typically lived three times as long as the older group.

When gene-expression studies were applied to these patients, distinct clusters of patients began to emerge, based on their tumor biology, how the tumors developed, and their responsiveness to chemotherapy. The tumor cells in older patients, regardless of their other biologic traits, were uniformly unresponsive to therapy; for these patients there is no biologic opportunity for the merciless 7 and 3 regimen to work. That, combined with normal age-related health changes and complications, says Rao, might be the reason for these patients' poor prognosis.

Rao is collaborating with other institutions to amass a total of 1,200 patient samples. "We already have drugs that target some of the overexpressed pathways found in older AML patients' tumors," she says, so if the initial results hold true, "we will be in a position to actually design clinical trials that are individualized," selecting new therapies to try based on the genomic makeup of each patient. At the very least, she says, physicians will be able to spare patients the risks and side effects of regimens like 7 and 3 when they are genetically destined to have no treatment benefit.

Pattern hunting

Guessing a tumor's destiny has become easier over the last 10 years. As techniques such as high-throughput sequencing have become more widely available, researchers have been able to create genetic sketches of one malignancy type after another—and in the class of liquid tumors, the subtypes of leukemia, lymphoma, and myeloma are legion.



Tannishtha Reya is seeking out the biologic weaknesses in cancer stem cells, which even the successful drug Gleevec can't kill.

"The idea is simple," says Sandeep Dave, pointing to a computer screen displaying a grid of jumbled green and red blocks. The grid maps the expression of 20 genes (a selection out of 25,000) in a group of patients with diffuse B-cell lymphoma—one of the most aggressive forms of lymphoma, and one of the first tumor types to be examined genomically. "Two samples of this type of tumor could look absolutely identical under the microscope and have two very different outcomes," explains Dave. "No one could understand why there was so much heterogeneity in these patients."

Genomic technology was able to turn the gene jumble on his screen into a very clear pattern. "A technique called hierarchical clustering groups together the samples with the most similar genetic makeup," he says, clicking to the next screen, where the jumbles are rearranged into a patchwork pattern—several patients who share a high-low expression pattern followed by several more who share a low-high pattern. The small sample of each patient's intricate genetic quilt shows that while no two tumors are exactly alike, there are definitely ways to classify them into similar groups. And the clinical outcomes of these groups could be predicted based on their genetic expressions.

When Dave applied this same technique to follicular lymphoma, the disease that struck his med-school mentor, he looked for gene expression patterns that might explain the disease's widely variable prognosis—and he found them, though not where he thought he would. "We found genetic signatures in the patients' immunologic makeup that are associated with survival," he says, and these signatures are more predictive of positive outcomes than any clinical factors such as age and stage at diagnosis.

This type of genomic profiling is taking place throughout the field of oncology, in solid and liquid tumors alike—Duke's Institute for Genome Sciences & Policy has already initiated clinical trials in breast, lung, and prostate cancers, all based on genomic research. Led by Joseph Nevins, PhD, Anil Potti, MD, and Phillip Febbo, MD, the trials use genetic profiling of tumors as a means of selecting chemotherapy for participating patients. But Dave says that the hematologic research goes beyond predicting which existing chemotherapy will work best. "Chemotherapies in nearly every combination have been

tested in almost every hematologic malignancy," he says. Instead, he believes that the patterns he finds can help focus research resources on the experimental models that are most likely to work. "There are over a hundred biotech companies trying to create new cancer drugs, in addition to the usual drug companies," says Dave. "So the question is, what molecules are most likely to have an effect on which types of liquid tumor?"

An army of trials

Dave's work to answer that question supports clinical investigators such as Anne Beaven, MD, who are launching an array of new, genomically based trials for liquid tumor patients. Beaven will lead the lymphoma trial at Duke that tests whether the genetic patterns found by Dave correlate to clinical response to therapy; if successful, another wave of trials will use these patterns to pair patients with treatments. "It's the first lymphoma trial of this kind at Duke," Beaven says, that takes a patient-by-patient approach. "These aren't going to be the therapies where 80 percent of all people with diffuse B-cell or follicular lymphoma will respond; we're looking for agents that will work in 80 percent of a particular selection of patients."

Beaven's goal is to have clinical trials open for every major type of lymphoma. "It's important, as an academic medical center, that we offer that kind of availability," she says. Oncologist Cristina Gasparetto, MD, a specialist in multiple myeloma, agrees. "At Duke we're trying to offer lots of options to patients and their physicians," she says, from proper staging of the disease to experimental approaches for high-risk or relapsed patients.

One option that is continually being improved at Duke is stem-cell transplant—currently the best treatment bet for many patients with multiple myeloma and other hematologic malignancies. Duke researchers Nelson Chao, MD, and Joanne Kurtzberg, MD, are among those who have made transplantation a feasible, survivable therapy for an ever-growing number of cancer patients in an ever-widening age range, but the process is still a toxic one, and often frail patients are not good candidates. Gasparetto is investigating ways to improve transplantation and expand the number of patients who are eligible for the procedure. "We are testing new, powerful pre-transplant therapies that have lower toxicity," she says, noting that these therapies are also being examined in patients who are not transplant candidates.

Last year Gasparetto was part of a Duke team, led by David Rizzieri, that published

Sandeep Dave's genomic tumor profiling goes where microscopes cannot, predicting with new accuracy how lymphomas will (or won't) respond to therapy.

Dan Wechsler examines six-year-old Marisa Rosa of Wake Forest, North Carolina, in the pediatric hematology clinic at Duke Children's Health Center.

Protecting young blood

SEVEN YEARS AGO, DAN WECHSLER, MD, PHD, treated a three-month-old girl who developed acute myelogenous leukemia (AML). She died within 12 days of her diagnosis. But her short life led to a discovery that may change the fates of other babies and children who develop AML.

Wechsler, who is chief of pediatric hematology-oncology at Duke, says that his lab discovered a genetic mutation in the baby's blood samples—a translocation of two genes that has since been confirmed in other pediatric AML cases around the world. Gene translocations are seen most commonly in children's cancers, but also play a role in other cancers: the Philadelphia chromosome that plays a role in chronic myelogenous leukemia (CML) in adults results from just such a mutation, and its discovery paved the way for the development of Gleevec.

The translocation Wechsler discovered is a swap of the usual location of two genes: CALM and MLL, a gene often involved in both pediatric and adult leukemias. With the help of researchers in his lab such as Catherine Lavau, DVM, PhD, who developed mouse-model techniques that are used worldwide in the study of leukemia, Wechsler is investigating how a disruption in CALM may trigger AML by interfering with a vital cellular process. As was the case in CML, such understanding of pediatric AML development could lead to improved, targeted therapies.

Pediatric hematologic malignancies, though they may have the same names as their adult counterparts, are actually different cancers in their genesis and in their behavior. And for the most part, they have much better outcomes: 50 percent of children who develop AML are cured, compared to only 20 percent of adults with AML; the cure rate for children with acute lymphocytic leukemia (ALL), the most common cancer in kids, is 80 to 85 percent in children and only 30 percent in adults. But childhood cure rates weren't always so good—in 1965 only 10 percent of children with ALL survived. Wechsler says this amazing advancement is a direct result of the Children's Oncology Group (COG), a National Cancer Institute-supported cooperative research group that links academic medical institutions throughout the country. Duke's pediatric oncology program is a member of the COG, and pediatric cancer patients at Duke can be enrolled in one of more than 65 current COG studies for a range of cancers—five of which are for leukemia.

All Duke pediatric hematology-oncology faculty members belong to the COG, and they make up a multidisciplinary team that combines pediatric oncologists, radiation oncologists, surgeons, pathologists, and social workers to treat children with cancer.

To learn about the program or any of the COG clinical trials, please call 919-684-3401.

the largest study yet to demonstrate the effectiveness of mismatched adult immune-system (blood) transplants. "If we look at the reasons that transplants fail patients," says Rizzieri, "it's because of either relapse, infection, or not having donor matches." And as the success of mismatched transplantation improves, "you broadly expand those who have a donor to include almost everyone," he says. "That, combined with a well-tolerated preparative regimen for transplant, would significantly decrease the toxicity of the approach—and allow us to offer this therapy to patients who otherwise wouldn't have a meaningful chance of cure."

A better watch-and-waiting game

For patients with aggressive, quickly fatal malignancies such as multiple myeloma, clinical trials can offer promise where previously there was none. For patients with less aggressive cancers, such as chronic lymphocytic leukemia (CLL), this new wave of genomic clinical trials helps refine both treatment options and the decision of whether to treat at all.

CLL is one of the most common leukemias, and according to several studies it is one of the most strongly heritable diseases period. Survival of CLL patients ranges wide, and in many cases patients can live for years before symptoms or disease progression mandates treatment. Oncologists Daphne Friedman, MD, and Mark Lanasa, MD, PhD, are building a CLL clinic at Duke that will both treat CLL patients and help sort out what genes are involved in the development of the disease.

From a research perspective, Friedman says that because CLL is such an indolent disease in many patients, it's an excellent model to study genetically. "Patients live for a long time, often without needing aggressive therapies. They come in again and again, so we can take repeated samples and look at how things change"—because the initial genetic trigger of CLL in a patient may not be what promotes tumor cell survival during the course of treatment or in cases of relapse. Understanding how the disease changes genetically will help physicians choose therapies wisely for CLL patients. "There are about five drugs that could be applied to CLL patients," says Friedman, "but there aren't a lot of ways to forecast which one—if any—is likely to have an effect." Also, she says, understanding changes in oncogenesis can have a significant impact in many other types of cancer.

Lanasa treats patients in the CLL clinic and also collects data—from both the patients and their families. He is part of a national collaboration gathering data from families with high incidence of CLL to help sort out what group of genes causes the cancer. Unlike cancer-causing gene mutations such as that of BRCA-1, which have a high "penetrance"—meaning that if you have the mutation you are highly likely to develop the disease in your lifetime—CLL seems more likely to be the result of an unruly set of low-penetrance genes. "There are probably five or 10 genes that are involved," says Lanasa, "but you need lots of families to figure this out for sure."

Lanasa and Friedman's work is a continuation of their fellowship research, which they both completed last year under J. Brice Weinberg. Their new clinical initiatives, like those of Dave, Beaven, Gasparetto, Rao, and Reya, illustrate how the journey of potential new therapies from laboratory bench to bedside is growing steadily shorter. For some patients this offers new hope for effective treatment. But Lanasa says that even for patients whose cancer is being followed but not treated, clinical studies can provide a much-needed mental benefit. "Watching and waiting can be frustrating; it can feel passive to people," he says. "When patients can participate in clinical trials, it's something active they can do about their disease." □

To learn more about Duke hematologic malignancy clinical care and current clinical trials, please call 919-684-8964.