HOW PERSONALIZED MEDICINE IS CHANGING: BREAST CANCER

Testing for dozens of biomarkers means oncologists can fine-tune breast cancer treatments in ways not even dreamed of just an eye-blink ago. ***By Heather Millar***

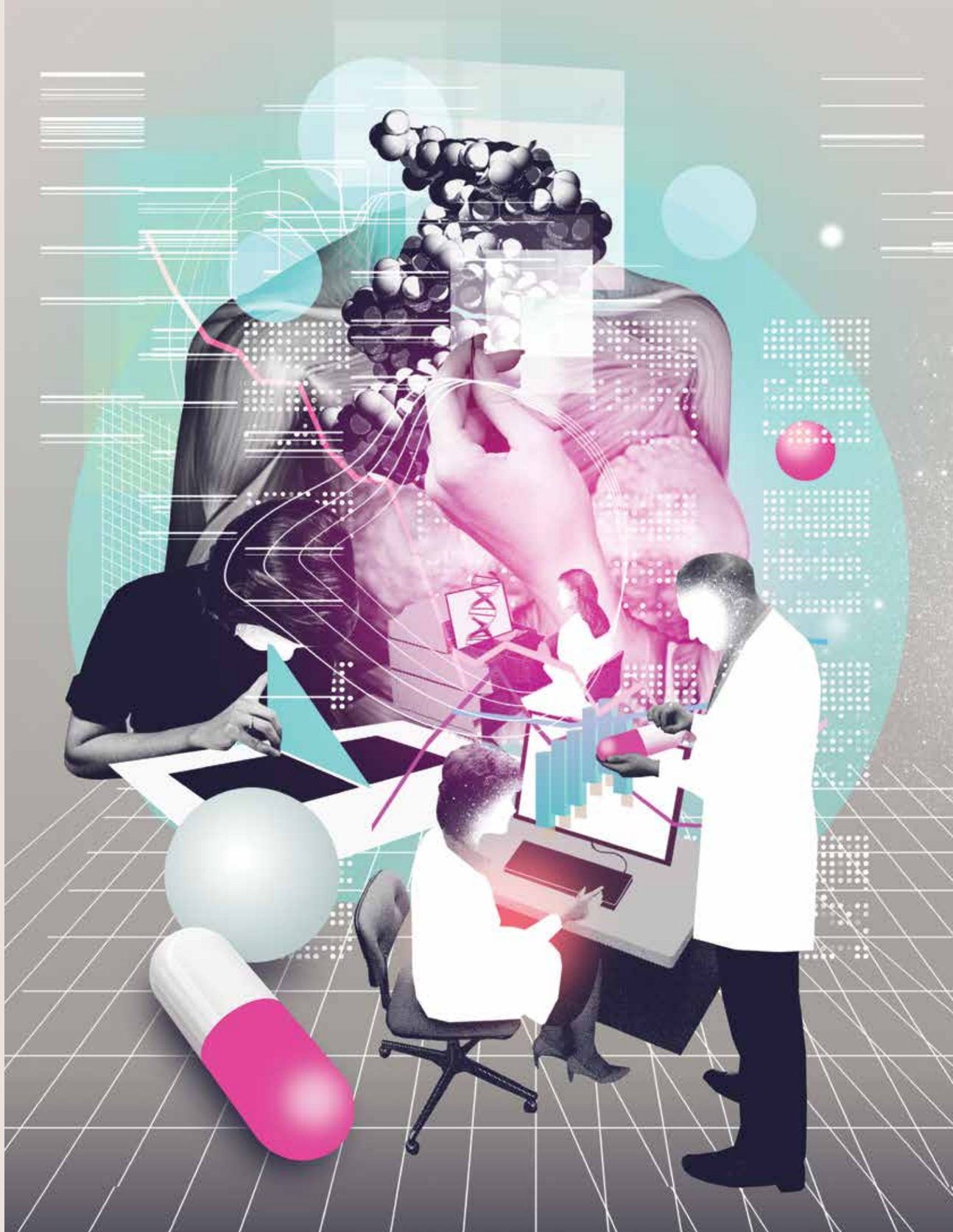

ILLUSTRATION BY MARIO WAGNER

In 2011, a 52-year-old runner and yoga enthusiast walked into the office of Monica Loghin, a neuro-oncologist at MD Anderson Cancer Center in Houston, complaining of numbness and weakness in her lower limbs and difficulty controlling her bladder. ¶ The symptoms were of grave concern, as the patient had previously undergone surgery for breast cancer that had spread to her brain. If such a cancer returns post-surgery, that is often a sign the patient doesn't have much time left.

An MRI confirmed that the breast cancer had again spread to the woman's cerebrospinal fluid. Loghin ordered testing of that fluid to see if the patient might have certain biomarkers that could be targeted by existing drugs. (A biomarker is a DNA sequence or protein associated with the disease; different biomarkers can suggest specific treatments, depending on the disease and other factors.) She asked for tests that could detect tumor cells circulating in the blood.

The cancer cells in the fluid bathing the woman's spinal cord and brain chambers did, in fact, have a lot of the protein that controls a glucose (sugar) transporter that drives cancer cells. The cancer cells in the fluid also had a lot of HER2, a protein associated with aggressive breast cancers but also treatable with a drug called Herceptin (trastuzumab). The drug is usually taken intravenously, but Loghin had heard of a couple of cases in which Herceptin was delivered directly into the cerebrospinal fluid via a flexible tube, or catheter. The patient agreed to this experimental treatment.

It took only a week for the news to improve. After the first infusion of Herceptin, the patient's cancer numbers were down. Within a few weeks, her cancer cell numbers had fallen so low that her immune system had begun to take over, clearing out the remaining cancer cells. Nearly two and a half years later, the patient is still alive and well enough to do yoga. Another MD Anderson patient who had a similar disease profile and therapy is also alive and well one year after treatment.

This case outlines the dream of personalized medicine: A disease is analyzed at the molecular level. The analysis identifies a drug target. The drug gets delivered where it needs to go. The patient gets better. And while this hopeful scenario has yet to become commonplace, it is becoming more and more the norm for many breast cancer patients.

As it relates to breast cancer, such a targeted approach to treatment is increasingly possible. "I am optimistic about personalized medicine," Loghin says. "I want to develop a protocol to identify receptors in the spinal fluid and blood that we can use for targeted therapy."

Even more tailored treatment is on the horizon. In the next few years, patients with breast cancer can expect increasingly detailed diagnostic tests, tests that may predict side effects of treatments like chemotherapy or radiation, and better guidance in choosing the drug, or drugs, most likely to eradicate their disease. Genomic discoveries may also help patients with advanced, aggressive cancers, a group for whom even the latest treatments often fail. In these ways and many others, breast cancer prevention, treatment, and care are a microcosm of the slow but inevitable shift in healthcare.

EARLY PROGRESS

All cancers are caused by genetic mutations that somehow confer an advantage on the cancer cells, causing — or simply allowing — them to grow out of control. Genes, the biological "operating system" that determines how living things grow and maintain themselves, can get garbled in a variety of ways. As cells divide, genes may get miscopied. Or they can be copied too many times. Or perhaps a bit of genetic code gets left out. It turns out that different breast cancers might have very different profiles of these genetic mistakes, affecting different groups of genes.

Even before the human genome was completely sequenced in 2003, researchers had sketched in some of the biological details that would allow them to differentiate one breast cancer from another.

"In terms of a targeted approach to breast cancer, how far do you want to go back?" says J. Leonard Lichtenfeld, deputy chief medical officer for the American Cancer Society. "For example, we've been measuring hormone receptors for decades."

In 1951, researchers discovered that the hormone estrogen could drive the growth of breast cancer. Fifteen years later, they found the "receptor" for estrogen, the part of the cell that locks on to the hormone. That gave doctors a "target," something that medications could be designed to block. Similar discoveries were made for the hormone progesterone. Nearly 75 percent of breast cancers are estrogen sensitive or progesterone sensitive or both, and drugs have been developed to interfere with the receptors for these hormones, thus slowing or stopping cancer growth or preventing a recurrence of the cancer.

In 1981, scientists identified a cancer-causing gene in mice. Four years later, they found the human version of this gene, HER2, the same gene that was overactive in the cerebrospinal fluid of the MD Anderson patient. Approximately 20 to 25 percent of breast tumors have this protein. A HER2-positive status used to mean a poor prognosis, but since the development of the genetically targeted drug Herceptin, patients with HER2-positive breast cancer actually benefit from some of the best treatment options.

While estrogen receptor, progesterone receptor, and HER2 tumor markers were helping target treatments in women diagnosed with cancer, progress was also being made identifying people at risk of developing cancer in the first place. In 1990, human geneticist Mary-Claire King showed that a location on chromosome 17 (subsequently identified by Myriad Genetics as BRCA1) was linked to many breast and ovarian cancers, proving wrong many who doubted the relationship between genetics and complex human diseases. Later in the 1990s, Myriad developed tests for BRCA1 and BRCA2, which both mark a susceptibility to breast cancer and ovarian cancer and tend to be hereditary.

But all of these advances — impressive and important as they were — simply identified single genes or proteins that might affect the course of a breast cancer case. The next breakthrough gave medical science a glimpse at our entire genetic blueprint, allowing for a global analysis and development of panels of genes for prognosis and targeted treatment of breast cancer.

"The good news is that breast cancer is one of the forms of cancer that has had a personalized approach for the longest time."

THE AGE OF GENOMICS

Have you ever held some sand in your hand and assumed the grains were mostly the same? Then have you looked at that sand through a magnifying glass and gasped at the diversity of colors, shapes, and origins, the bits of glass, of shell, of stone, of bone, every hue of the rainbow? That's one way of understanding how our vision of breast cancer is changing thanks to genomics.

Once the entire human genome was sequenced in 2003, researchers and companies began to think about more complete genetic profiles of diseases like breast cancer. New prognostic tests soon followed. In 2006, researchers published pivotal results showing that the Oncotype DX test that measured estrogen-sensitive patients' risk of a breast cancer recurrence based on a panel of 21 "oncogenes," or genes that affect cancer cells, could also predict the degree of benefit a patient could expect from chemotherapy. In 2007, a 70-gene panel called MammaPrint won FDA approval. Both tests genetically analyzed a patient's tumor tissue, resulting in a score that predicted the risk of the cancer recurring. A low score meant the cancer probably wouldn't come back; a high score meant recurrence was likely. This allowed doctors to determine which patients might not need chemotherapy and which patients did.

These developments that gave doctors tools to "stratify," or categorize, different cases of breast cancer are part of the oncology-ward argot. You can't sit in the waiting room of a care center without hearing snippets of conversation that include genetic and molecular information. "ER/PR status," "HER2 positive," "BRCA1 and BRCA2," Oncotype DX and MammaPrint scores — all these terms punctuate the patois of the breast cancer ward.

That's partly because genomics is poised to make treatment even

more hopeful for breast cancer patients as clinicians and researchers discern more genetic details that drive the disease — and that affect how patients respond to treatment. Doctors hope that better targeting will help patients whose disease has spread to other organs, or who relapse many years after the primary cancer — patients for whom there are now few options.

NEW INSIGHT, NEW DRUGS, NEW HOPE

It often surprises people to learn that a cancer's DNA is different from the DNA of the person who has the cancer. Often, the malignant DNA becomes wildly different. If it weren't a crazy genetic outlier, it probably wouldn't be cancer.

That reality led to the launch of The Cancer Genome Atlas (TCGA), a project of the National Cancer Institute that in 2009 brought together researchers from many disciplines to compile comprehensive genomic maps of 20 common cancers, including breast cancer. In the United Kingdom, the Breast Cancer Somatic Genetics study hopes to create genomic profiles of 500 breast cancer cases.

Just a decade ago, this kind of mapping would have involved a multistep process that cost millions and would have taken years or decades. Now, "next-generation" gene-sequencing technology has made it possible to analyze the genome of a breast cancer tumor with a one-step process for just thousands of dollars. This has accelerated our understanding of how breast cancers differ molecularly from one another.

"The most exciting thing that's happening now is the ability to do tumor sequencing," says Kelly Marcom, who started a hereditary breast cancer clinic at Duke University School of Medicine in the late 1990s. "It will be a while before we know what to do with all that information and how to marry that with the patient's own genetics and how to use that to refine treatment. But that's where the field is exploding."

"There's so much information at a raw data level," says Gavin Gordon, vice president at CollabRX, a San Francisco-based company that is trying to make the avalanche of genomic data understandable for doctors and patients. "Think of it this way: There used to be 50,000 academic papers on cancer annually. Now there are 100,000. There used to be just a few cancer drugs. Now there are about 500 in development."

Sequencing of tumor genomes has revealed several subtypes of breast cancer. In 2012, a paper published in the journal *Nature* reported results from the genomic sequencing of 510 breast tumors in 507 patients, part of the TCGA project. In all, the sequencing found 30,626 mutations in the cancer cells, which could be roughly divided into four groups:

- "Basal-like" tumors resemble the deepest layer of skin and account for a small, deadly percentage of cancers, often called "triple negative." These cancers appear to be entirely different from other breast cancers, actually sharing some features with a type of lung cancer and with ovarian cancer.
- Two related subtypes — "luminal A" and "luminal B" — include hormone-sensitive cancers. Doctors have struggled to understand why some of these cancers react to hormone-targeted drugs and others don't. Now doctors think that perhaps luminal A patients might do well with just hormone therapy while luminal B patients need more aggressive treatment such as chemotherapy.
- HER2-enriched cancers also turn out to fall into two groups, perhaps explaining why not all of these cancers react to the HER2-targeted drug Herceptin.

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"With the new tools, it's like a new microscope," explains University of North Carolina Medical School Professor Charles Perou, one of the *Nature* study authors and leader of the TCGA Breast Cancer Working Group. "Looking at the cancer DNA landscape, it's a 10,000-fold leap. We're learning a huge amount."

One of the biggest surprises, Perou says, is that molecular differences and biological pathways may connect cancers that we didn't even know were related. In other words, it's not where the cancer emerges that may always be the most important. Rather, it's how the cancer cells work. That's why there is a movement toward reclassification of cancers based on their molecular profile versus traditional classification based on tissue of origin.

In addition to the connection that Perou's team found between basal-like breast tumors and some ovarian and lung tumors, other teams have found that certain breast cancers may share features with bladder cancer. A European team found that some breast cancers share regions of "hyper-mutations," overactive garbled DNA, with some types of lung and blood cancers.

The realization, thanks to genomic research, that where cancers start may not be as important as their molecular profile has helped researchers to think outside the box. Many new studies are exploring whether drugs thought useful for only one kind of cancer might work for cancers that arise in a variety of organs.

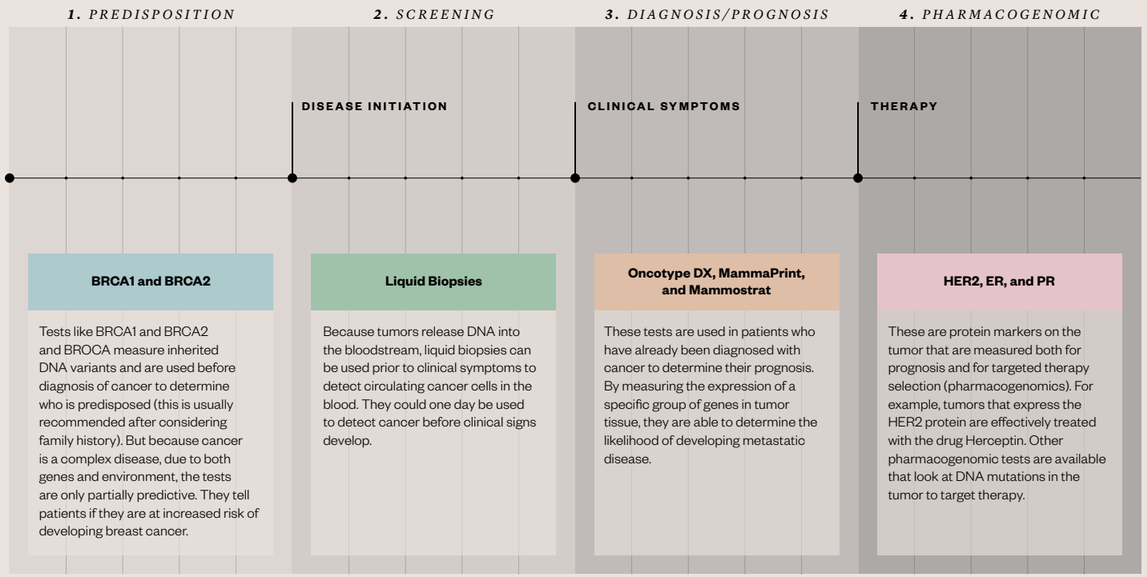
For instance, a November 2013 study combined Nexavar (sorafenib), currently approved to treat kidney and liver cancer, with Stivarga (regorafenib), approved for colorectal cancer, and a class of drugs called PI3K/AKT inhibitors. The researchers found this to be effective for a variety of cancers, including breast cancer.

Another study, presented at the 2013 Breast Cancer Symposium in San Antonio, showed that combining standard anti-hormone therapy Femara (letrozole) with Sprycel (dasatinib), a drug approved for chronic myeloid leukemia, doubled progression-free survival time in patients with advanced breast cancer. One of the ways that Sprycel works is by blocking a protein called Src. This protein has recently been implicated in the spread of breast cancer to bones.

In April 2013, the FDA awarded a similar combination — Femara plus an experimental Pfizer drug called palbociclib — "breakthrough therapy" status, meaning the drug's approval will be expedited. (The

The Testing Timeline

At which point in the clinical course of a disease are various genomic tests used?



combination met the primary goal of its phase 2 trial in February of this year.)

While there is still a lot of work to be done to translate this new knowledge into tests and treatments for patients, other studies have begun to pinpoint which genetic markers give clues as to which drugs might best fight a certain cancer. For instance, a January 2013 review by Perou and Washington University School of Medicine Professor Matthew Ellis summed up the results of six other genomics studies, which identified several genetic mutations that might make promising targets for new therapies.

"A new treatment paradigm is rapidly evolving," Perou and Ellis wrote in the review, published in *Cancer Discovery*. "Deep genomic analysis will drive treatment decisions based on ... cell type and pathway-matched therapies."

Every month, it seems, a team identifies a new possible target mutation. As breast cancer specialist and Stanford University Medical Center Professor George Sledge put it in a presentation at the Personalized Medicine World Conference in January, "It turns out that we don't need a magic bullet to cure cancer. We need a magic shotgun."

MORE TESTS, BETTER TESTS

Until the new discoveries lead to clinical research that makes truly personalized breast cancer medicine possible, the most immediate result of the genomic revolution has been the proliferation of new tests based on next-generation sequencing. Agendia, the company that pioneered MammaPrint, has now developed the Symphony Breast Cancer Genomic Profile to yield an even more detailed picture of each case. After the Supreme Court last year ruled that Myriad Genetics could not patent the BRCA1 and BRCA2 genes, the company announced that by 2015 it would replace its BRCAnalysis test with a broader myRisk panel that analyzes 25 genes associated with hereditary cancers, including breast, colorectal, ovarian, endometrial, pancreatic, prostate, gastric, and melanoma.

Several competing companies have started marketing BRCA tests, including Invitae, Ambry Genetics, and Quest Diagnostics, and the courts are likely to be busy for years with patent challenges and counterchallenges. At $1,500, Invitae's BRCA test is a less expensive alternative to Myriad's BRCAnalysis, and the company also offers a High-Risk Hereditary Breast Cancer panel that goes beyond BRCA testing and includes other genes associated with high-risk breast cancer syndromes. In addition to BRCA testing, Ambry Genetics offers BRCAplus and BreastNext, the latter of which looks at 16 genes implicated in hereditary breast cancer. And Quest's BRCAvantage tests for genetic risk of hereditary breast and ovarian cancer.

Beyond prognostic tests, women with early-stage cancer have other tools to gauge their risk of recurrence, like the recently FDA-approved Prosigna test from NanoString, an analysis of 50 genes. For patients with HER2 results that are unclear, the new TheraLink test may help clarify which drugs might be most effective. Another genomic test, called the RD-100i OSNA system, can detect even "micro-metastases" to a patient's lymph nodes, usually the first place that breast cancer spreads. A Washington University team is working to pinpoint patients who may test HER2 negative but nonetheless have an activating HER2 gene mutation that may affect their case.

Even more fine-grained tests are coming. A team at Purdue University is building on genomic information to flag cancers by how they process certain proteins. Other teams are working on genomic tests to identify "cell-free cancer DNA" — i.e., cancer DNA circulating in the bloodstream — as a way to monitor for remaining cancer cells that cannot be seen by conventional imaging methods, and to monitor response to treatment. These tests, which detect circulating tumor DNA, are often referred to as liquid biopsies. Janssen Diagnostics' CellSearch system is the only test of this kind so far to receive FDA clearance.

What all this means for patients now is that ever more information is becoming available to help guide decisions about treatment. Armed with genomic information, breast cancer patients may now feel comfortable forgoing certain treatments that might not be necessary for their unique case.

"We are at the beginning of a revolution," says the American Cancer Society's Lichtenfeld. "Breast cancer, as with many others — lung, melanoma, etc. — has a number of therapies in the pipeline, and that number is increasing dramatically. What does the future hold? Some successes, some failures. Will it lead to a cure? I can't say that. Will it lead to control of breast cancer? That's a real possibility."

BETTER CHOICES FOR PATIENTS

When a person is first diagnosed with cancer, the first panicked thoughts run along the lines of a medical sledgehammer: "Cut it out! Nuke it! Poison it!" Unfortunately, because cancers turn out to be so multifarious, "doing everything" doesn't always guarantee survival. Not only that, but aggressive treatments like chemotherapy and radiation may come with costs and side effects that can sometimes be as devastating as the original cancer.

Ann Meredith, a mother of two who lives outside Philadelphia, says she "completely flipped out" when she was diagnosed with breast cancer in August 2013. She says she was ready to try anything and consulted three doctors within a week, seriously considering chemotherapy, radiation, and a double mastectomy. The third doctor, Massimo Cristofanilli at Jefferson University Hospital, suggested that Meredith have her tumor analyzed before she went the scorched-earth route.

Cristofanilli sent her biopsy sample to Agendia, which analyzed Meredith's tumor for hundreds of gene signatures. The results showed her cancer to be a luminal type: estrogen sensitive and low risk. Meredith opted for breast-conserving surgery, or lumpectomy, as well as radiation and hormone therapy.

"I really felt that because this information was available, it wasn't a cookie-cutter approach to cancer treatment," Meredith says. "It wasn't an emotional reaction to the fear. It was based on the best science available."

Obviously, this science will evolve. But some hopeful developments are likely to hit the clinic soon. Researchers are making gradual progress in understanding how best to treat aggressive and metastatic cancers. Genomic information is leading to new combination therapies for aggressive triple-negative and metastatic HER2 cancers. For example, one study showed a benefit to combining a traditional chemo drug, carboplatin, with a new DNA-damaging agent called a PARP inhibitor.

Oncologists have long known that combinations of drugs often work better than one drug alone. In a 2011 study, women with hormone-sensitive cancers (estrogen and/or progesterone positive) took a standard hormone-blocking drug, Aromasin (exemestane), in combination with Afinitor (everolimus), a drug that inhibits a protein called mTOR. This combination seemed to slow the progression of advanced cancers.

Recent research has focused on which drugs are best to partner with Herceptin, one of the first genomics-based drugs for breast cancer and one that targets a gene that directs the production of HER2.

> "When I first heard I was hormone negative and HER2 positive, I thought, 'I'm dead.' But this treatment targeted my tumor type. In breast cancer, personalized medicine is moving quickly and slowly at the same time. In my case, I won the cancer lottery."

At the annual San Antonio Breast Cancer Symposium last December, researchers reported that, after surgery, pairing Herceptin with the chemo drugs docetaxel and carboplatin appeared to be the best option. Another study presented at the symposium provided early evidence that combining Herceptin and Tykerb (lapatinib), another HER2-targeted drug, might work better than Herceptin alone. The researchers found that 84 percent of the patients who received this combination remained cancer free, compared to 76 percent who received Herceptin alone.

Another study found that for women with metastatic disease, giving Herceptin and an intravenous treatment called Perjeta (pertuzumab), which binds to a different part of the HER2 protein, slowed the progression of their cancer.

Others are working on genomic signatures that might help predict which patients will react badly to chemotherapy or radiation treatment. Inform Genomics has created a test to assess which patients are most at risk of nausea and vomiting and is developing a platform that will also assess the risk of four more common side effects: mouth sores, fatigue, difficulty thinking clearly, and nerve problems. A team at the University of Manchester in the United Kingdom is studying genomic signatures of long-term side effects of radiation treatment.

Many companies are finding genomic signatures to pair with particular drugs (aka "pharmacogenomics"). BioMarin in Northern California hopes to use genomics to predict which patients will benefit most from treatment with PARP inhibitors. Los Angeles-based Arno Therapeutics is working to understand which patients might benefit from drugs that block the activity of progesterone receptors. Teams

at the University of Michigan and Sloan-Kettering Cancer Center are both working to find markers that might signal resistance to hormone therapy. As well, companies like Foundation Medicine and resources like My Cancer Genome are working to tie treatment to a deeper understanding of genomic changes that contribute to disease.

NEW SYSTEMS FOR TRIALS

While teams the world over are turning genomic discoveries into advances for patients, many researchers say the system for clinical trials needs to change. The current process tests potential treatments one at a time in "double-blind" trials, in which one group of patients gets the new therapy and a control group does not. Trials of new drugs go through three or four phases, checking first for safety, then whether it works or not, and finally tracking long-term effects. The process takes years.

But our growing understanding of cancer genomes is suggesting many, many possible new drug targets. We're also able to retest things that were rejected because they didn't work on enough people but might work if they were targeted to patients with the right genomic profile. Researchers say these probably need to be tested in combination, all at once, in a process that doesn't take decades. For instance, the NeoALTTO study, published in February 2012, showed that two anti-HER2 drugs in combination seem to work better than just one or the other.

The FDA is trying to adjust to these new realities by creating new procedures that allow drugs to be tested more quickly: Breakthrough Therapy is a new program announced in June 2013 that builds on previous programs such as Fast Track, Accelerated Approval, and Priority Review that might be used for therapies based on biomarkers.

Many companies and academics — Novartis, Amgen, and the Open Medicine Institute, to name just a few — are experimenting with new models for clinical trials. The one that is probably furthest along is the I-SPY trial of breast cancer patients conducted by the University of California at San Francisco and enrolling patients at medical centers around the country. In this study, researchers are testing multiple drugs from multiple companies all at once. In I-SPY, breast cancer patients agree to have systemic treatment before surgery, so that researchers can see how their tumors react. Depending on results, drugs can be added or dropped.

"The goal of the I-SPY trial is to quickly find the best drugs for each person and for us to really understand how to tailor treatment," explains Laura Esserman, principal investigator for the trial and director of the Carol Franc Buck Breast Care Center at UCSF.

The trial, which has completed phases 1 and 2, drops therapies that don't seem to work and "graduates" therapies that seem to have great benefit. This has already happened for drugs that seem to affect triple-negative or basal-like cancers and HER2 cancers. A third phase will begin this year.

Anne Marie Hallada, a 41-year-old mother of four from Palo Alto, California, felt a lump in her breast while nursing her youngest son, now 2. She enrolled in the I-SPY trial and started chemotherapy with a combination of Taxol (paclitaxel) and an experimental drug. After three weeks, the doctors took an MRI image of her breast to prepare for a biopsy. They couldn't find the tumor.

Arriving at the hospital for a chemo infusion the next day, Hallada met her oncologist in the hospital elevator. "She was jumping up and down, she was so excited by what had happened," Hallada says. Just to be safe, Hallada completed her chemo regimen and then had surgery. She opted not to have radiation but will have Herceptin infusions through this spring.

"In surgery, they just found a crater where the cancer had been," Hallada says. "When I first heard I was hormone negative and HER2 positive, I thought, 'I'm dead.' But this treatment targeted my tumor type. In breast cancer, personalized medicine is moving quickly and slowly at the same time. In my case, I won the cancer lottery."

WHAT'S NEXT?

Fifteen years ago, hormone receptors — those that made a patient estrogen or progesterone positive or negative — were the only real molecular targets for diagnosing and treating breast cancer in a personalized way. Now breast cancer patients can benefit from information on a variety of genomic markers: HER2, AKT1, PIK3CA, PTEN, and "kinase" genes that cause DNA to get rearranged or overexpressed in a way that leads to cancer. They can benefit from earlier and more detailed diagnosis and monitoring, thanks to genomic tests that build on our growing knowledge. But we're still in the very early days of truly personalized medicine, experts say.

The genes, gene interactions, and environmental factors that may drive or affect a case of breast cancer are dizzyingly complex. For instance, a study published in *Nature* in May 2012 analyzed the breast cancers of 100 patients. The researchers were able to identify "driver mutations" — that is, genetic changes that seemed to drive the cancer to grow — in about 40 different cancer genes. Some patients had only one driver mutation, but others had as many as six drivers. Neither pharmaceutical research nor genetic research nor federal regulation is set up to target six things at once. That means the disease will probably continue to surprise us for years or decades to come, researchers say.

Why do some cancers react to certain drugs and not to others? Why might a cancer become resistant to a drug? How can we predict these outcomes and changes? How do we choose a therapy that is just right for a particular patient? These are the kinds of questions researchers will try to answer in coming years.