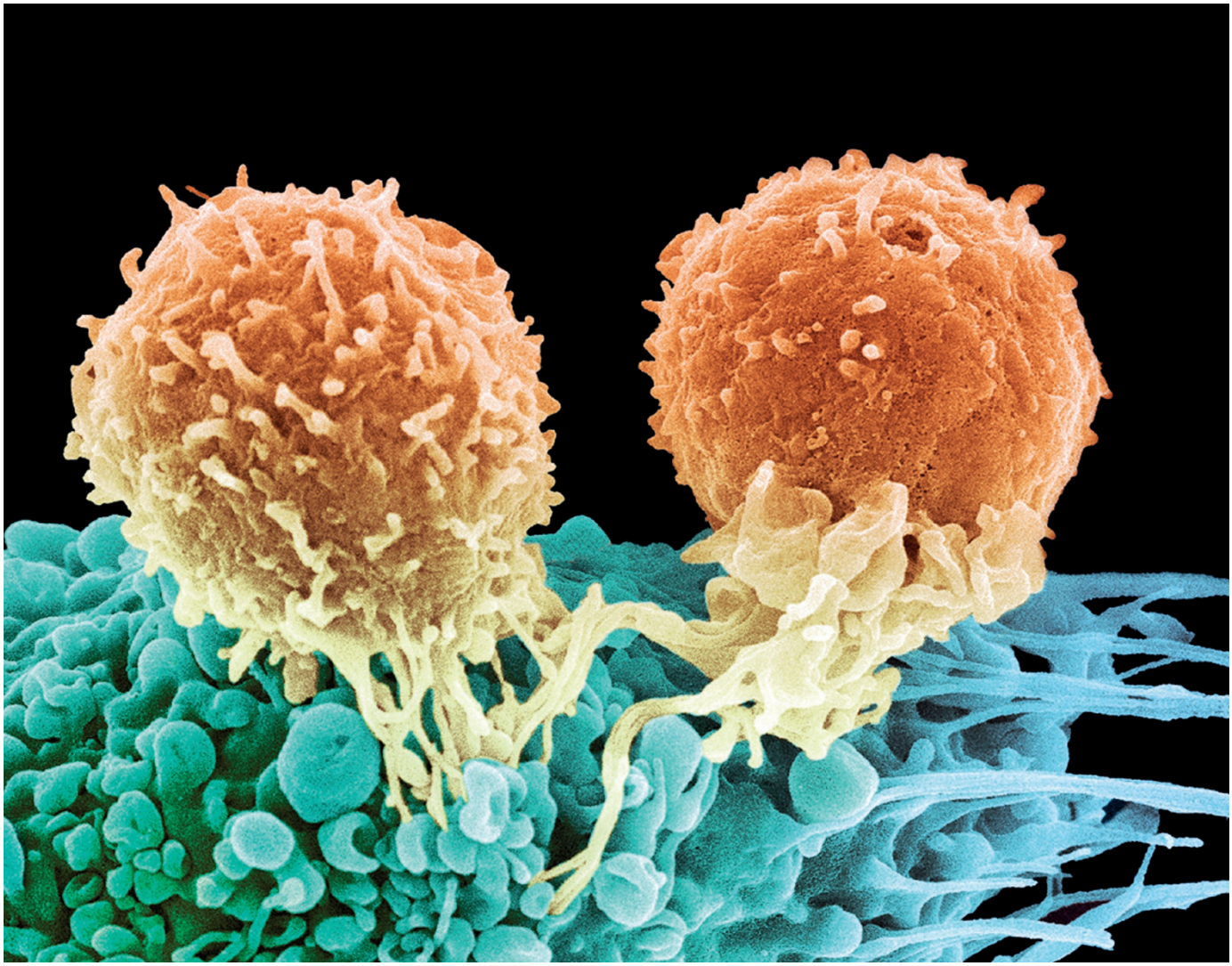

STEVE GSCHMEISSNER/SPL

White blood cells called T cells recognize and attack cancer cells as part of the immune response, which is boosted during immunotherapy.

IMMUNOLOGY

Another shot at cancer

Targeting the immune system to fight breast cancer was all but dismissed in the 1990s, but the strategy is making a big comeback with the possibility of a breast-cancer vaccine.

BY CHARLES SCHMIDT

While on her hospital rounds at the Walter Reed National Military Medical Center in Bethesda, Maryland, Elizabeth Mittendorf encountered a patient whose story is still fresh in her mind 14 years later. The woman had been successfully treated for breast cancer more than 15 years earlier, but the disease had returned. "And I wondered, how is it possible that someone beats breast cancer only to face it again?" recalls Mittendorf, now a surgical oncologist at the MD Anderson Cancer Center in Houston, Texas. "To me this could only mean that this woman's immune system had failed her."

Mittendorf has since dedicated much of her career to breast-cancer immunotherapy — a field that is just starting to hit its stride. Immunotherapy drugs boost the body's inflammatory response against malignant tumours. None have been approved for the treatment of breast cancer, and many uncertainties remain, but this is an undeniably exciting time. As of August, more than 40 clinical trials of breast-cancer immunotherapies are underway worldwide, and two of them are in phase 3 — the final stage before regulatory approval can be sought. People with breast cancer already benefit from effective treatments, and 5-year survival rates for newly diagnosed cases top 90% in the United States. But drugs that enhance the immune system's battle against malignancy might prevent recurrences altogether, says Mittendorf, the principal investigator in a phase 3 trial of a vaccine called NeuVax.

"What we hope is that immunotherapy will someday cure

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breast cancer," says Mary Disis, an oncologist at the University of Washington School of Medicine, Seattle. "The immune system remembers cancer antigens, and it can seek out and kill off metastases anywhere in the body, including in the bones and the brain. We just have to figure out how to sustain that response before it's exhausted."

KIN MAN HUI/SAN ANTONIO EXPRESS-NEWS/ZUMAPRESS.COM

Trials of breast-cancer vaccines are underway, but the vaccines may work best when combined with other treatments.

A LONG HISTORY

The concept of cancer immunotherapy dates back more than a century. The bone surgeon William Coley, who worked at what later became the Memorial Sloan Kettering Cancer Center in New York, injected his patients with a killed bacteria vaccine during the late 1800s in the hope of stimulating the body's defences. During the 1990s, physicians began treating people with cancer with high doses of interleuken-2 (IL-2) and interferon-γ (IFNγ) — inflammatory cytokines released by infection-fighting white blood cells called T cells. Some people with cancer have lived for decades with the help of cytokine treatment, but because the inflammation that high-dose cytokines generate is systemic there can be life-threatening side effects, including vascular leakage and kidney damage.

A crucial breakthrough came in 1996, when James Allison, an immunologist at MD Anderson Cancer Center, and his colleagues showed that it was possible to amplify anti-cancer immunity by taking the brakes off a molecular checkpoint that would otherwise dampen the immune response[1]. The body relies on these checkpoints to regulate inflammation and limit the risk of autoimmune disease. But as they deliver this essential service, checkpoints interfere with the immune system's efforts to destroy growing tumours. Allison's research showed that blocking a checkpoint known as CTLA-4 located on T-cell surfaces enhances the immune response to cancer with fewer side effects than those brought on by IL-2 and IFNγ.

In 2011, the US Food and Drug Administration (FDA) approved the CTLA-4 inhibitor ipilimumab for use in treating advanced melanoma. During phase 3 testing, people with the disease who were treated with ipilimumab lived an average of four months longer than those who went without the drug[2]. Some super-responders are still alive today.

While Allison targeted CTLA-4, other researchers were exploring the clinical possibilities of another immune checkpoint on T cells: programmed cell death protein-1 (PD-1). PD-1 binds to its ligand on cancer cells forming a complex called PD-1/PD-L1 and hiding the tumours from the immune system. Preventing the formation of these complexes has proved beneficial in cancer treatment. In December 2014, the PD-1 inhibitor nivolumab became the latest immune checkpoint therapy to gain FDA approval, specifically for the treatment of metastatic lung cancer[3]. European approval followed in April.

At first, there was widespread doubt that checkpoint inhibitors would be any use against breast cancers. The approach only works in tumours that have already been invaded by tumour-targeting white blood cells called tumour-infiltrating lymphocytes (TILs). Melanoma and lung tumours contain a lot of TILs, which makes them easy targets, but breast cancer tends to have relatively low levels of TILs. "So the thinking was that it wouldn't respond as well to immunotherapy," says oncologist Leisha Emens at the Johns Hopkins University School of Medicine in Baltimore, Maryland. "And since breast cancer was already being treated with effective drugs, it wasn't associated with unmet medical needs in the same way that melanoma and lung cancer were."

As checkpoint inhibitors make their way onto the market, attitudes have clearly shifted. "The entire medical community is awakening to an appreciation of their potential role in treating all cancers," says Jill O'Donnell-Tormey, chief executive officer and scientific director of the Cancer Research Institute, a non-profit organization based in New York. "This is why you're seeing all these clinical trials in breast cancer now."

TRIAL BY VACCINE

Of the more than 40 ongoing breast cancer immunotherapy clinical trials monitored by the Cancer Research Institute, roughly two-thirds involve vaccines. Breast-cancer vaccines take a number of different forms: NeuVax, for instance, is derived from the cell-surface protein HER2, which some breast tumours have in large quantities, and is a target for the drug Herceptin (trastuzumab). Vaccines are also made from cancer-cell DNA, or entire cancer cells, and in some cases they are custom-made from a patient's own white blood cells exposed to tumour antigens in the laboratory.

Whatever their origin, cancer vaccines are designed to stimulate a particular kind of anti-tumour immunity, specifically: type 1 immunity. Type 1 immune responses depend on CD4 T-helper cells that secrete highly inflammatory cytokines, such as IFNγ and tumour necrosis factor-α (TNF-α). In turn, these cytokines activate the CD8 T cells that go on to attack and kill cancer cells.

But vaccines may not work well enough as breast-cancer treatments by themselves. During the phase 1/2 trial of NeuVax, for instance, 89.7% of treated women achieved 5-year disease-free survival compared with 80.2% of women who did not receive the vaccine[4] — a result that some found discouraging. Mittendorf, however, argues that NeuVax was able to cut what would have been a 20% risk of 5-year recurrence in half, "which is a fairly impressive number that's certainly of interest to patients."

A phase 3 trial, called the PRESENT study, will randomize 700 women with early-stage breast cancer and low to intermediate HER2 expression to receive either NeuVax or an immune-stimulating chemical called granulocyte-macrophage colony-stimulating factor.

According to Mittendorf, if the trial reaches its endpoint of 3-year disease free survival, the FDA will consider the vaccine for approval — results are expected in 2018.

An increasing number of researchers, however, believe that the future of breast-cancer immunotherapy lies in giving vaccines and checkpoint inhibitors as combined treatments. In this way, vaccines will stimulate T-cell responses in the breast that checkpoint inhibitors can then amplify and sustain. Discussions about combining NeuVax with checkpoint inhibitors in future trials are ongoing. "I think this will be an efficacious strategy," Mittendorf says.

HITTING THE TARGETS

Meanwhile, the field is grappling with how to match people with breast cancer with the appropriate immunotherapy. The degree to which women with the most common form of breast cancer, oestrogen-receptor positive, will benefit from immunotherapy remains an open question, according to Carsten Denkert, a physician at the Institute of Pathology, Charité University Hospital in Berlin. Many oestrogen-receptor positive tumours, which make up 80% of all new diagnoses, are slow growing and respond well to existing hormonal treatments, such as tamoxifen or aromatase inhibitors.

Mittendorf points out that the number of women who die of oestrogen-receptor positive cancer that has stopped responding to existing therapies exceeds the number of women diagnosed with more aggressive types of breast cancer. The "challenge and opportunity," she says, is to make oestrogen-receptor positive breast cancers better candidates for immune therapy, for instance, with combination treatments.

According to Christopher Heery, director of the Clinical Trials Group in the Laboratory of Tumor Immunology and Biology at the National Cancer Institute in Bethesda, Maryland, most immunotherapy trials look at highly aggressive triple-negative tumours, a much needed focus given that these cancers lack receptors for oestrogen, progesterone and HER2 — targets of existing cancer drugs. Debu Tripathy, who chairs the Department of Breast Medical Oncology at MD Anderson Cancer Centre, points out that the more mutated antigens that cancer cells carry, the more foreign they look to the immune system. And since triple-negative tumours express more mutated antigens than other breast cancer types, he says, they could be especially good candidates for immunotherapy.

Researchers hope to identify biomarkers that can help to predict which women with breast cancer will respond best to immunotherapy, but reliable candidates remain elusive. Checkpoint blockades in breast cancer have for the most part been limited to drugs that inhibit PD-L1. Examples include MPDL3280A, an engineered monoclonal antibody manufactured by Basel-based Roche and currently in phase 1 testing for triple negative breast cancer, and pembrolizumab, manufactured by Merck in Kenilworth, New Jersey, which is in phase 2 trials with the same purpose in mind. It was initially thought that better responses would correlate with high PD-L1 expression levels; so much so that clinical trials have excluded women with breast cancer shown to be PD-L1 negative on screening. But PD-L1 expression is dynamic and varies not just between individuals, but also over time. Up- and downregulation of PD-L1 by cells is a normal response to excessive inflammation — cells upregulate it to reduce inflammation and downregulate it when the inflammation subsides.

Levels of PD-L1 may vary then depending on when samples are taken, and researchers still debate whether low PD-L1 levels should affect study enrolment. "Some people will say 'You need high PD-L1 for checkpoint inhibitors to work and others will say 'we ran a study and PD-L1 didn't matter,'" Disis says. "The fact is that it's just not a great biomarker." According to Heather McArthur, a medical oncologist at Memorial Sloan Kettering Cancer Center in New York, there are other promising possibilities, including a marker for T-cell activation called ICOS, which predicts better responses to ipilimumab in patients with melanoma and a marker for T-cell proliferation called K167.

Another valuable biomarker, says Denkert, could be the amount of TILs in the tumour. oestrogen-receptor positive cancers tend to have low TIL levels, but that is not necessarily true of more aggressive malignancies, such as triple-negative and HER2-positive breast cancer. According to Denkert, about 25% of all aggressive breast cancers are "lymphocyte predominant," meaning that the number of TILs exceed the number of malignant cells. Another 25% of cancers have no TILs whatsoever, and the remaining 50% sit somewhere in between. Denkert's research shows that a breast tumour's TIL count is predictive of the response to chemotherapy — a high count predicts better responses — and he expects it to do the same for immunotherapy. "Tumours with zero lymphocytes will have only a small chance of responding to checkpoint blockade," he says.

"What we hope is that immunotherapy will someday cure breast cancer."

Boosting otherwise small amounts of TILs using vaccines could be a winning approach, Denkert says. But he thinks that in some instances, tumours characterized by low TIL levels might remain intrinsically invisible to the immune system even with this treatment because they do not express enough T-cell receptors. Denkert now plans to investigate that hypothesis in an upcoming clinical trial sponsored by the German Breast Group, a network of the country's academic research institutions.

Heery, however, cautions that not all TILs are equal. "Characterizing them is just as important as counting them," he says. TILs could reflect type 1 immunity or type 2, which can suppress anti-tumour responses, Heery says. That is a crucial distinction, assuming that, as some research suggests, there tends to be a disproportionate number of type 2 TILs in breast cancer. Denkert agrees, but adds that approaches to discern type 1 and type 2 lymphocytes in tumour samples are in development. "Immunologists will say 'the immune system is complicated and we have to look at the different types of immune cells,' which is completely true," Denkert says, "but we pathologists see this type of characterization as a second step that follows an initial effort to quantify the overall number of immune cells in the tumour. If we combine both worlds and accept different approaches, we can generate a more complete picture."

As is the case with other experimental treatments, immunotherapies are being tested mainly in advanced, metastatic breast cancer. "Drug development is always done in the metastatic setting first to try to find the agents that work rapidly," Heery says. The problem with cancer immunotherapies — especially vaccines — is that they can take up to three months to build up an adequate response, "so if sceptics don't think they're working well enough, it could be that we're just testing them in the wrong setting."

Ultimately, immunotherapies may have more success when used to treat early-stage breast cancer, and McArthur is one of the few investigators working in that setting. She selectively breaks tumour sections into fragments by freezing them with a tool that looks like a biopsy needle — the tiny tumour fragments are thought to attract a more robust immune response to cancers that are not highly immunogenic to begin with, she says. She is now testing this cryoablation approach in combination with ipilimumab in women with newly diagnosed oestrogen-positive cancer, independent of their HER2 status.

"This is an incredibly exciting time to be in oncology," she says. "We're seeing remarkable advances with immunotherapy in other solid tumours like melanoma and lung cancer, and I'm enthusiastic we'll have success in breast cancer too. We're on the cusp of a new era." ■

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