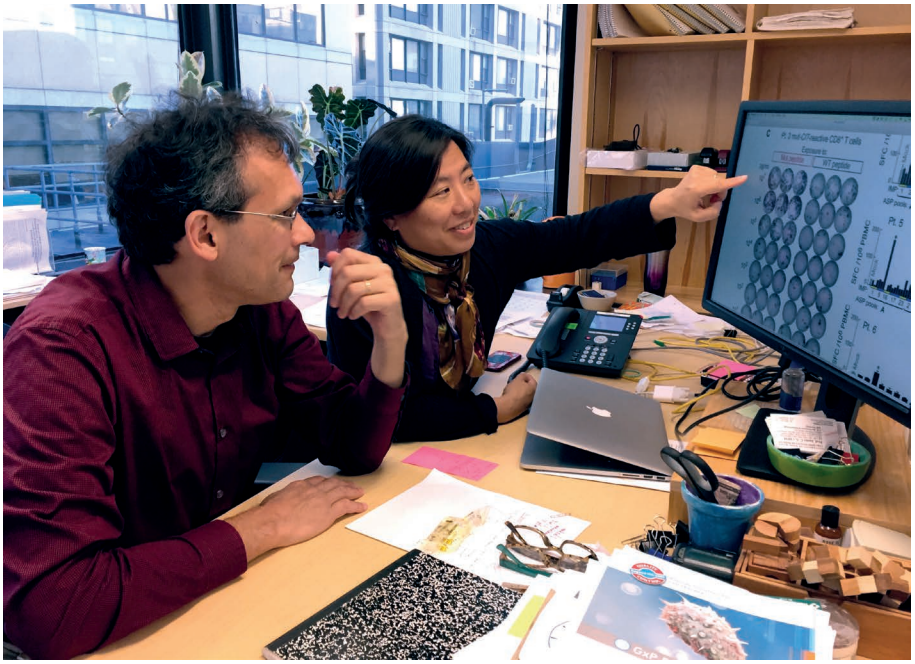
CANCER VACCINES

# Calling cancer's bluff

*Neoantigen vaccines exploit state-of-the art tumour-genome sequencing and analysis to provide uniquely personalized immunotherapy.*



GIACOMO OLIVEIRA

**Catherine Wu and Nir Hacohen looking at an image of T-cell responses.**

**BY SARAH DEWEERDT**

Cancer is famous for its ability to deceive, appearing to the immune system as normal tissue while wreaking destruction on the body. But what if cancer cells had 'tells' — subtle but unmistakable characteristics that revealed their true nature?

A growing number of scientists say that neoantigens, which are peptides (fragments of proteins) found only on the surface of cancer cells, could be those tells. They are working to develop vaccines that use neoantigens to help patients' own immune systems to fight tumours.

Neoantigens arise as a twist on the body's normal process of distinguishing 'self' from non-self. As part of this process, peptides called antigens are normally displayed on the outside of the cell. Cancer cells are highly prone to developing multiple mutations and, by chance, some of these mutations alter the amino-acid sequence of the peptides, converting them from normal antigens to neoantigens.

But when cancer cells adorn themselves with neoantigens, they are advertising that they don't belong — showing their hand as surely as the flicker of an expression across a card-player's face. A single base-pair change to a DNA sequence, and a single amino-acid difference in the resulting protein, can be enough to alert the immune system that something is amiss, and cause it to mount a response to the tumour.

Three independent studies over the past four years have reported on the experience of roughly two dozen people with melanoma, who were given experimental vaccines containing cocktails of neoantigens. After receiving the vaccines, all the participants made immune cells known as T cells that could specifically target their cancers.

Most remarkably, each person received a unique set of neoantigens. The researchers analysed individual tumour genomes and the patients' immune systems to make an optimal mix of antigens.

"It's really the first truly personalized medicine," says Nir Hacohen, director of the Center for Cancer Immunotherapy at the Massachusetts General Hospital in Charlestown, and a senior author on one of the studies.

"These early proof-of-principle studies have really provoked the field," says Nina Bhardwaj, an immunologist at the Icahn School of Medicine at Mount Sinai in New York, who has just begun a phase I clinical trial of a neoantigen vaccine.

Bhardwaj and others caution, however, that much larger studies will be needed to confirm the effectiveness of the neoantigen approach. Dozens of tricky details would then need to be worked out before vaccines are ready for wide use. Identifying the neoantigens made by an individual's tumour, and then predicting which of those molecules the patient's immune system might recognize, remains enormously challenging.

## CANCER-ONLY CLUB

Neoantigens "have always been conceptualized as very important, but we just didn't have the tools before," says Catherine Wu, an oncologist at the Dana-Farber Cancer Institute in Boston, Massachusetts, who collaborates with Hacohen.

One reason that neoantigens have long been considered promising targets is that they are unique to cancer cells. By contrast, other antigens that have been explored for cancer immunotherapy can also be expressed in normal cells, making healthy tissues potentially vulnerable to an immune response. And because neoantigens appear only when cancer develops, the immune system has a better chance of recognizing them in the first place. "The beauty of this vaccine approach is the ability to target just the tumour cells," says Robert Schreiber, professor of pathology and immunology at Washington University School of Medicine in St Louis.

In the 2000s, Schreiber's group laid some of the theoretical groundwork for neoantigen vaccines with mouse studies that showed how the immune system's response to neoantigens drives the evolution of cancer cells, ultimately enabling the tumour to escape from immune control. The idea of a vaccine is to strengthen the body's response to neoantigens it already recognizes, and to teach it to target neoantigens that it didn't respond to before.

The current excitement about neoantigen vaccines represents the confluence of several advances. Next-generation DNA sequencing technology has made it feasible to rapidly scan the 30 million base-pair protein-coding regions of both tumour and healthy cells and identify discrepancies that may give rise to neoantigens. Increasingly sophisticated computer algorithms are helping to identify the best neoantigens to include in a vaccine. And advances in manufacturing make it possible to quickly produce small quantities of diverse molecules for individual vaccines.

The advent of another form of immunotherapy — checkpoint inhibition, which stops tumours from suppressing immune-system

activity — has also contributed to making vaccination feasible.

Clinical researchers see neoantigen vaccines and checkpoint inhibition as complementary treatments that might often be used together, especially for patients with large tumours or metastatic disease. "The neoantigen vaccine is like the steering wheel, to guide the immune response," Hacohen says. "The checkpoint blockade is removal of the brakes."

## VALIDATING VACCINES

The first human study of a neoantigen vaccine for melanoma, published in 2015 (ref. 1), involved just three participants. It was led by oncologist Gerald Linette, at the Washington University School of Medicine, who is now at the University of Pennsylvania in Philadelphia. The patients made T cells that targeted the neoantigens they had been given in the vaccine, but the study included little clinical data. Two other small studies, both published in July 2017, provided the first hints that neoantigen vaccines might alter the course of a patient's disease.

In the study on which Hacohen and Wu collaborated[2], six people with melanoma received vaccines containing as many as 20 neoantigens each. Four of the patients were cancer-free two and a half years later. The tumours also disappeared in the other two patients, but only after treatment with a checkpoint inhibitor.

The other 2017 study[3], led by Ugur Sahin at Johannes Gutenberg University of Mainz in Germany, involved 13 people with melanoma, whose vaccines each contained a mix of 10 neoantigens. Nine of these people were cancer-free after a follow-up period of 12–23 months: eight had been given the vaccine alone and the other an additional checkpoint inhibitor.

Each of the three trials used a different method of delivering the neoantigens. Linette's study used dendritic cells, which have a key role in presenting antigens to the rest of the immune system. The team filtered precursors of these cells from each patient's bloodstream, then matured them in the laboratory and exposed them to synthetic peptides mimicking mutations in the patients' tumour-cell genomes. The peptide-loaded dendritic cells were then returned to the patients using an intravenous infusion.

The two studies published this year took a more conventional vaccine approach. Wu and Hacohen's group delivered neoantigen peptides. There is a long history of using peptides in vaccines, and it is easy to switch out different peptides to make an individual cocktail for each patient.

Sahin and his collaborators used RNA molecules encoding neoantigen peptides in their vaccines. RNA, like peptides, is fast and flexible, and it has an extra advantage: "It is its own adjuvant," Sahin says, referring to an ingredient that is added to a vaccine to help stimulate an immune response. As RNA is the genetic material in many viruses, the body tends to be on high alert for it; the mere presence of certain forms of RNA can kick-start the immune system.

Despite these diverse approaches, the vaccines all had effects. "So that's the good news," says Schreiber, who was not involved in the studies. "But there's not been enough treatment yet to know, is any one better than the other?"

The only way to resolve this, say Schreiber and other scientists, is through more studies with larger numbers of patients. Such studies are also necessary to confirm whether neoantigen vaccines actually help people to live longer.

## RAISING RESPONSE

All studies of neoantigen vaccines in people published so far have involved melanoma. It's thought to be a good target because melanoma cells tend to have a large number of mutations.

Lots of mutations means lots of neoantigens, which are needed because patients do not produce T cells corresponding to every neoantigen in a vaccine cocktail. In Sahin's study, the overall response rate was 60% — enough to make a difference to patients, but leaving plenty of room for improvement.

Until researchers get better at predicting which neoantigens will generate a response, they need to target cancers with a high mutation rate, says Fred Ramsdell, vice-president of research at the non-profit Parker Institute for Cancer Immunotherapy in San Francisco, California. As well as melanoma, that category includes lung, stomach, colorectal and cervical cancers. "We want to be really efficient in generating these responses before we go to tumours with a mutation load that's relatively low," Ramsdell says.

Despite this caution, clinical trials are tackling cancers that carry very few mutations, such as the brain cancer glioblastoma, as well as others with moderate mutation rates such as breast cancer. The results may eventually reveal whether there is a certain mutational threshold required for neoantigen vaccination to be feasible.

*"The neoantigen vaccine is like the steering wheel, to guide the immune response."*

Running in parallel is work to improve the prediction of which neoantigens will provoke an immune response. Algorithms are constantly being refined. The Parker Institute has launched a project to coordinate this research that involves pharmaceutical, biotechnological, academic and non-profit partners.

Researchers are watching to see how close the project can come to reaching a 100% response, says Bhardwaj, whose lab is participating. "Can you predict well enough that everything you pick is immunogenic?"

To reach that level, scientists will have to nail down many other technical details such as the dose and timing of vaccination and the best adjuvant. They will have to learn how to combine neoantigen vaccines with checkpoint inhibitors and other therapies, and how the microenvironment — the immediate area around a tumour — affects the function of the immune system. And they will have to make vaccines much more quickly — in several weeks or less rather than the three months or so it took in the recent clinical studies.

Academic labs and biotechnology companies seem eager to take on the challenge. Hacohen and Wu are co-founders of Neon Therapeutics of Cambridge, Massachusetts, and Sahin is founder and chief executive of Mainz-based BioNTech, both of which aim to commercialize neoantigen vaccines.

Neoantigen therapy is seen as a promising treatment for cancer, and there are high hopes that the strategy will fulfil its potential. But there is also the legacy of cancer therapies with theoretical promise that haven't panned out. The neoantigen approach, says Wu, "is very rational on paper. But we have to just see how it works out in the clinic." ■

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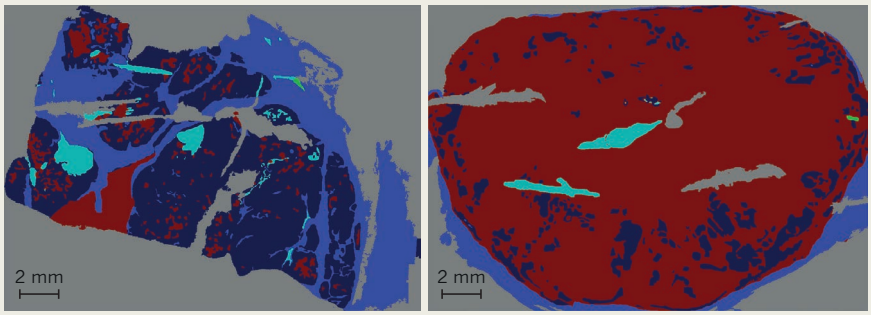
1. Carreno, B. M. *et al. Science* **348,** 803–808 (2015).
2. Ott, P. A. *et al. Nature* **547,** 217–221 (2017).
3. Sahin, U. *et al. Nature* **547,** 222–226 (2017).

## VISUALIZING A NEOANTIGEN VACCINE AT WORK

Computer visualization of metastatic melanoma cells, before (left) and after (right) the patient was treated with a neoantigen vaccine. After vaccination, about three-quarters of the cells were seen to be dying (red).




SOURCE: SAHIN, U. *ET AL. NATURE* **547,** 222–226 (2017)