to fight off the virulent Georgia 2007 strain.4 Antibodies alone are not fully effective,” Dixon says.

Over the past few decades, researchers have begun to understand why. Studies suggest that pigs rely heavily on killer T cells—and potentially other immune cells—to fend off ASFV, and stimulation of T cells can only occur if living viruses infect host cells.5 Only then are viral peptides processed and presented via cell surface receptors to T cells. This doesn’t happen with the dead viruses traditionally used in the vaccine experiments.

Recognizing this, researchers developed vaccine candidates with live, but weakened, forms of ASFV. They took advantage of the fact that many ASFV strains have mutated over time, becoming less aggressive to domestic pigs and their wild relatives. In 2019, a group of Spanish researchers injected a number of domestic pigs with a weak strain of ASFV genotype 2 that had been isolated from a wild boar captured in Latvia. The vaccine caused mild, transient symptoms involving fever and joint swelling in some animals, but they all survived after being exposed to pigs that carried the virulent genotype 2 strain Georgia 2007.6

As researchers have amassed more knowledge about ASFV’s biology, they’ve adopted a more targeted approach in attenuating viruses by removing specific genes that make it so deadly. As Dixon puts it, the goal is to strategically disarm the virus so the porcine immune system has a chance to develop long-lasting antibodies and to prime T cells to attack the virus. In 2016, for example, her group created a single-engineered strain of ASFV genotype 1 by knocking out eight genes and interrupting two genes the virus uses to dampen pigs’ interferon type 1 response, a path that helps curtail viral replication. All five animals immunized with this approach in attenuating viruses by removing specific genes that make it so deadly. As Dixon puts it, the goal is to strategically disarm the virus so the porcine immune system has a chance to develop long-lasting antibodies and to prime T cells to attack the virus. In 2016, for example, her group created a single-engineered strain of ASFV genotype 1 by knocking out eight genes and interrupting two genes the virus uses to dampen pigs’ interferon type 1 response, a path that helps curtail viral replication. All five animals immunized with this approach survived a challenge with a virulent genotype 1 strain.

Dixon’s team has achieved similar success with other gene deletions6—7 and, with funding from the Biotechnology and Biological Sciences Research Council in the UK, is working with a New Jersey-based biotechnology company to create a viable vaccine candidate for the Georgia 2007 strain using this approach. Meanwhile, at Plum Island Animal Disease Center, microbiologist Manuel Borca has developed four gene-deleted vaccine candidates that promote animals against the same strain—each carrying one, two, or three deletions.8 A number of other groups, including one in China, are also working on similar gene-deletion approaches.

Although they’re effective, safety is still a significant concern. Some of Dixon’s immunized pigs developed a slight fever, which most veterinary vaccine companies would consider an unacceptable safety risk, though further gene deletions and modifications have greatly reduced or eliminated this fever while maintaining good efficacy, she says. Part of the challenge in identifying the right genes to remove to strike that balance between safety and efficacy is ASFV’s unusual genomic complexity.

Like some other DNA viruses, ASFV has a very large genome—this is about 190 kilobases,” notes Mwangi, making it longer than the RNA genomes of Ebola, HIV, Lassa, Marburg, and rabies viruses put together.

Another major barrier in developing live vaccine candidates is that they’re difficult to produce in bulk. ASFV replicates longer than the RNA genomes of Ebola, HIV, Lassa, Marburg, and rabies viruses put together.

Instead, researchers have to continuously harvest fresh macrophages from animal blood or other body tissues.

“You will never get a uniform, reproducible [vaccine] product” this way, remarks Volkan Revilla of the Spanish National Research Council’s Center for Molecular Biology “Severo Ochoa” in Madrid. Finding a cell line that lasts is “one of our most important objectives at the moment.”

Protein cocktails

To get around these issues, researchers such as Mwangi are trying another strategy: a subunit vaccine. Viral vectors—such as adenoviruses—are engineered to express cocktails of ASFV antigens. Once the vector abounds, stimulation B cells and T cells

A VACCINE HUNT

Researchers have tested these main approaches to develop a vaccine candidate for the ASFV strain that is currently killing pigs throughout Asia.

VACCINE STRATEGY #1: INACTIVATED VIRUSES

The traditional approach involves killing or inactivating viruses—for instance, through UV irradiation—so that they’re no longer virulent but retain viral antigens that stimulate the production of protective antibodies.

EFFICACY: These vaccines stimulate an antibody response in pigs, but they don’t protect against intact forms of ASFV. Researchers think this is because inactivated viruses don’t activate killer T cells.

SAFETY: Based on limited studies, no side effects have been shown so far.

COMMERCIAL PROSPECTS: Researchers have abandoned this approach because of the shortfalls in efficacy.

VACCINE STRATEGY #2: LIVE VIRUSES

Injecting tamer forms of virulent viruses could potentially stimulate antibody production and the all-important T cell responses without killing vaccinated animals.

EFFICACY: Both gene-deleted and naturally attenuated forms of ASFV stimulate the immune system to generate antibodies and killer T cells and usually offer protection against virulent genotypes of ASFV.

SAFETY: Vaccinated pigs can develop mild to debilitating symptoms, from fever to joint swelling.

COMMERCIAL PROSPECTS: Researchers are both testing ASFV strains that have naturally attenuated over time and genetically modifying virulent forms of the virus by removing sequences that code for harmful proteins. Scientists have yet to find a stable cell line capable of generating live vaccine candidates in bulk, but these types of vaccines are expected to be the first to hit the market.

VACCINE STRATEGY #3: SUBUNIT VACCINES

A third approach involves genetically engineering viral vectors such as adenoviruses to express combinations of ASFV antigens. Inside the body, the vector-encoded antigens are produced in the absence of the pathogen.

EFFICACY: Injections provoke the production of antibodies and killer T cells, but don’t seem to protect pigs against virulent forms of ASFV.

SAFETY: Vaccinated animals typically experience few or no side effects.

COMMERCIAL PROSPECTS: Researchers are testing different antigen combinations. Many consider this to be the preferred strategy for developed countries, although it’s expected to reach the market much later than live virus vaccines. Subunit vaccines can be easily synthesized in bioreactors and rapidly generated in bulk.