Pigs were the source of the 2009 H1N1 influenza pandemic.

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AGRICULTURE

Flu on the farm

Farms help to spread influenza but they might be an early warning system for the next human pandemic.

BY CASSANDRA WILLYARD

In December 2014, virologist Hon Ip received a shipment from a biologist in Washington state. It was a package containing nine dead birds.

Ip's job at the US Geological Survey's National Wildlife Health Center in Madison, Wisconsin, was to work out what had killed the birds. He was worried that it might be avian influenza. There had been an outbreak in South Korea earlier that year, and in December a novel version of avian influenza was detected in Canada, just 70 kilometres north of where the birds now in Ip's possession had been found. He feared that these waterfowl might also have been infected.

The cause of death was indeed avian flu. Whole-genome sequencing revealed[1] the presence of a highly pathogenic strain of the influenza virus. Such viruses do occasionally arise in the United States but this strain differed from all those that had been detected previously: it came from Asia.

For more than a decade, Ip had been monitoring wild birds for signs of Asian bird flu but had never found the virus. Now, less than a year after the virus emerged in China and South Korea, it had made the leap across the Bering Strait into the United States. "It is the scenario we'd been watching for since 2005," Ip says.

Over the next six months, the virus evolved in a variety of ways, jumped from wild birds to turkeys and chickens, and wreaked unprecedented havoc on the US poultry industry. More than 50 million chickens and turkeys in the United States were killed, either by the virus or by efforts to stop its spread, making this the largest and most expensive avian influenza outbreak in the United States.

Modern farms are particularly vulnerable to devastation from influenza. A large farm might hold tens of thousands of chickens or thousands of pigs in the name of efficient protein production, and this creates an opportunity for viruses such as influenza to mutate and spread.

But there is an even greater fear: that these ever-changing viruses will give rise to the next human pandemic. Last year marked the 100-year anniversary of a pandemic that killed as many as 50 million people worldwide. "We're worried," says Ip, "about another Spanish flu." To prevent that from happening, researchers need to bolster surveillance efforts and curb the spread of flu in animals.

THE BIRD FLU

There are four types of influenza. The most common, influenza A, can infect both humans and animals. Virologists classify these viruses into subtypes based on two proteins on their surface, haemagglutinin (H) and neuraminidase (N). There are 18 recognized haemagglutinin types and 11 neuraminidase types. The dead birds that Ip examined were infected with the H5N8 virus.

But viruses do not stay neatly in their assigned categories. "Flu viruses have an infinite capacity to mutate," Ip says. "They mutate at some of the fastest known rates" of any virus. They also change through a process called reassortment. The influenza A virus has eight RNA segments, and if more than one virus infects a single cell, the viruses can swap some of those RNA segments. This could give rise to an entirely new virus for which no human or animal has immunity, Ip says, and it is this

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constant shuffling that makes influenza so difficult to treat — and so dangerous.

The concern around avian influenza began in the late 1990s when a highly pathogenic strain of H5N1 began infecting people in Hong Kong. Until then, avian influenza had caused only mild disease in humans. But H5N1 was different. The first 18 cases in Hong Kong resulted in 6 deaths. On that occasion, there was no pandemic — no more human cases emerged. But in 2004, the World Health Organization (WHO) warned that the next pandemic could result in the deaths of up to 7 million people worldwide.

Health officials feared that deadly Asian viruses such as H5N1 might make the leap to North America, so Ip and others began monitoring wild birds for signs of such viruses. For nearly a decade, every search came up clean.

Then, in 2014, those nine dead birds arrived at Ip's lab. The moment the H5N8 virus crossed the Bering Strait and entered North America represented the dawn of a new reality. "Not only was it an exchange of an avian influenza virus, it was an exchange of a deadly form — a highly pathogenic virus," says David Swayne, laboratory director of the Southeast Poultry Research Laboratory of the US Department of Agriculture (USDA) in Athens, Georgia.

Another concern is that avian influenza viruses of Asian origin often have higher morbidity and mortality rates in humans than other avian flu strains, says James Kile, an influenza specialist at the US Centers for Disease Control and Prevention (CDC) in Atlanta, Georgia.

The H5N8 strain has not yet caused disease in humans but other avian virus strains have. In 2013, a new strain of avian influenza emerged in China: H7N9. Unlike the virus that caused the US outbreak, H7N9 did not typically kill poultry, at least not initially. Indeed, it caused such mild illness that it was not detected until it began infecting people.

To combat the spread of the virus, the authorities in China began closing live poultry markets in provinces where human infections had occurred. But these measures to curb the spread of influenza may not always have had the intended effect[2]. Rather than shutting all the markets at once, the closures happened at different times in different provinces. In Jiangsu, for example, the policy took effect in December 2013, whereas the neighbouring province of Anhui took no action until February 2014. This meant that although the measure seemed to work initially, poultry farmers in infected areas were able to send their birds to markets in neighbouring provinces that had not yet been affected, thereby spreading the virus.

The CDC currently ranks H7N9 as the influenza virus with the highest potential pandemic risk. The virus has made more than 1,500 people ill and killed at least 615 since 2013. But the threat seems to have abated, at least for the moment. During the winter of 2016–17, H7N9 evolved into a highly pathogenic strain. The Chinese government responded by mandating that poultry producers immunize their birds with a vaccine targeting both the H5 and H7 strains. The strategy worked. By June 2018, the vaccine had been linked[3] to a 92% decrease in H7 detection rates in poultry and a 98% reduction in human cases.

A CAULDRON OF VIRUSES

Some researchers are more worried about pigs than poultry. Gregory Gray, an epidemiologist at Duke University in Durham, North Carolina, considers pigs to be ideal mixing vessels for influenza viruses because the animals are susceptible to not only swine flu, but also avian and human influenza. Even so, flu viruses in swine often go undetected and unreported. "Influenza A viruses are largely tolerated because they don't cause a big problem, at least not in the pigs," Gray says.

The World Organisation for Animal Health, the Paris-based intergovernmental body that sets standards for reporting animal disease, requires that certain strains of avian influenza be declared. But pork producers do not need to report swine flu to the authorities.

In April 2009, officials in the United States detected a new strain of influenza in humans known as H1N1. The virus became known as swine flu and seemed to be the product of a reassortment between three viruses circulating in pigs. The virus spread quickly around the world, and two months later the WHO declared that the outbreak had reached pandemic status. In the wake of this pandemic, the USDA launched a programme in concert with industry and the CDC to conduct voluntary surveillance for swine flu. The goal is to keep tabs on the viruses that are circulating in pigs.

> ***"The picture we have of the types of viruses that are circulating is very superficial."***

Despite this, "the picture we have of the types of viruses that are circulating is very superficial," says Gray. That is true not only for the United States but also China, which is the world's largest producer of pork.

"There's a massive transition in China from small and medium-sized farms towards large industrialized farms, but we still see rather poor biosecurity," Gray says. When he and his colleagues toured farms in China, they noticed that personal protective equipment is used only sporadically, barriers to stop rodents entering are rare, and pigs are sometimes housed near ducks, geese or chickens. "It's a cauldron of virus mixing," Gray says.

In 2015, Gray and his colleagues launched a five-year study to examine the transmission of swine influenza in large pig farms in China. Results from the first year of that study[4] suggest that swine flu is fairly common in pigs and that farm workers are also being infected. The team found similar H1N1 viruses in pigs, workers and on surfaces in the barns.

Gray and other researchers are hopeful that improvements in technology will allow them to keep better tabs on influenza in animals and curb the spread of the virus.

STOPPING THE SPREAD

China has been vaccinating poultry against avian influenza but the practice is not common in the United States. No birds at all were vaccinated during the 2014–15 outbreak. According to Joelle Hayden, a spokesperson for the USDA's Animal and Plant Health Inspection Service, vaccination would be used only as part of an eradication effort for highly pathogenic strains of avian influenza, not as a replacement for eradication.

But vaccination can be problematic. Any virus that is not wholly eradicated could still mutate enough to render the vaccine against it ineffective. Even when an effective vaccine is available, its use is not guaranteed. A 2018 study[5] found that some H7N9 viruses had become lethal in ducks, yet only about 30% of China's duck population had been vaccinated.

Jürgen Richt, a veterinary microbiologist at Kansas State University in Manhattan, says that producers need something they can easily apply *en masse*, rather than injecting each bird individually. Richt and his colleagues are developing a sprayable live vaccine that protects against both avian influenza and the virus that causes Newcastle disease — another serious infection that affects poultry. So far, they have tested versions aimed at eradicating the H5, H7 and H9 strains of influenza. Richt is also working on a universal vaccine for humans that might eventually be used for animals too.

Richt and his colleagues have also created a pig that is genetically resistant to swine flu. This might protect not only the pigs, but also humans. Even if the pig can still be infected, its resistance to influenza could mean that it spreads less readily. But whether the US Food and Drug Administration (FDA) will allow such pigs into the food supply chain is not yet clear. "This is the biggest question at the moment," Richt says. So far the FDA has approved only one genetically engineered animal for food use: a salmon that has been modified to grow faster.

Even if these strategies are widely adopted, Ip emphasizes that we must stay vigilant. Another influenza pandemic is inevitable and no one knows exactly what it will look like.

"We always hone a strategy towards the last outbreak that we experienced," Ip says. But strategies used during the last outbreak may not work next time. "Never be dogmatic," he says. "The flu virus changes all the time." ■

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