

ON BORDER PATROL

The United States has embarked on a huge effort to try to track the H5N1 avian flu virus in birds migrating into the country. But is surveillance more urgently needed elsewhere? **Erika Check** reports.

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On a Sunday afternoon in June, graduate student Brad Comstock is standing on the flat, soggy tundra at the edge of the Bering Sea. He surveys the network of shallow ponds stretching out in front of him, and the vast delta reaching to the horizon beyond. Nestled down along the shores of the ponds are dozens of large, migratory sea ducks called eiders. Each bird is settled on a down-lined nest holding eggs that are just days away from hatching. The air is filled with the sound of eiders honking to each other over the constant, whistling wind. "This is eider heaven," Comstock says.

But he is about to stir up trouble in paradise. Carrying a fishing net and a backpack full of test-tubes and cotton swabs, he creeps towards one of the nests. At the last moment, he hurls the net towards a mother eider. She jumps to evade him, but too late; her wings flap angrily as the net ensnares her.

Comstock rushes in, extracts the offended bird and turns her upside down. He runs a cotton swab across her cloaca to take a faeces sample, then drops the swab in a test-tube. The sample will be held briefly at a nearby camp, then shipped to the National Wildlife Health Center in Madison, Wisconsin, where technicians will test it for the H5N1 avian influenza virus.

The 7.7 million hectares of the Yukon Delta National Wildlife Refuge in Alaska may seem like heaven to eiders. But to the US government, it is the frontline in the battle to protect the country from the deadly bird flu virus.

Comstock is part of a massive effort to track the possible entry of H5N1 into the United States. Since 2003, the virus, a more lethal strain than the flu viruses that normally infect birds, has rampaged through the rest of the world. More than 200 million poultry have died of H5N1 or have been culled to prevent its spread since 2004; 132 people have died after catching the virus from close relatives or directly from birds. So far, the virus hasn't learned how to jump efficiently from person to person. But public-health officials fear that if it does, a pandemic could follow, killing millions of people.

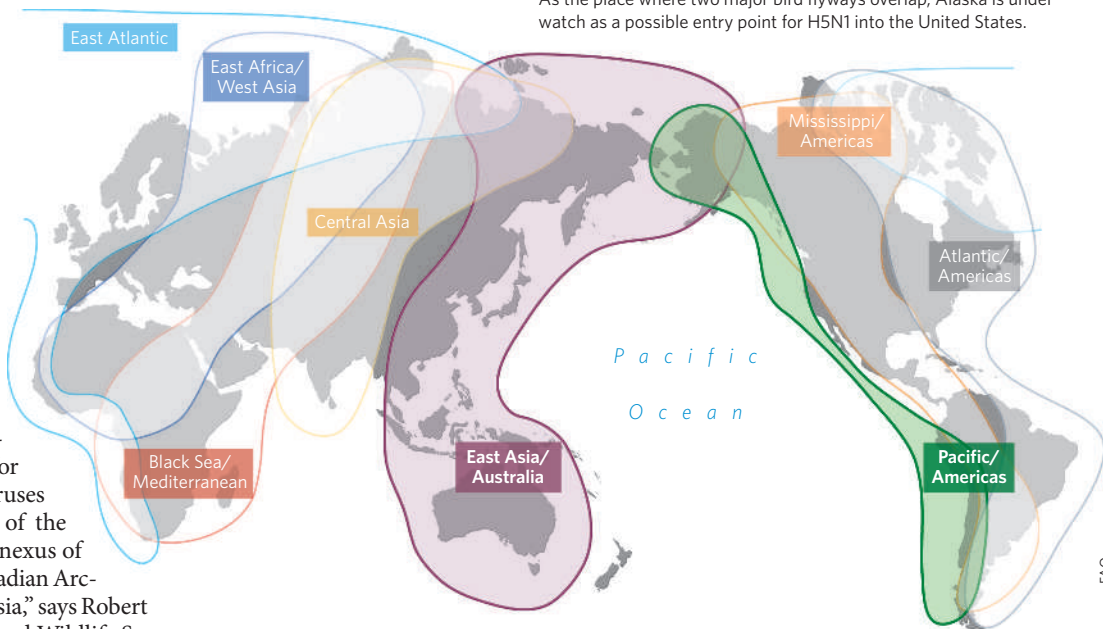
The US government is worried about wild birds because the country is linked directly to Asia — where H5N1 first appeared — through two overlapping migratory bird flyways (see map). Every year, birds such as pintails, eiders, ducks, godwits and geese cross from Asia over the Bering Sea into Alaska. These birds mingle with other migrating groups at breeding and wintering grounds in Russia and western Alaska. No one knows which of them might be



Brad Comstock is helping the US mission to test 15,000 wild birds for the flu virus in Alaska alone.

A ROUTE FOR FLU?

As the place where two major bird flyways overlap, Alaska is under watch as a possible entry point for H5N1 into the United States.



carrying H5N1, or whether the virus might leap from such a carrier into people or into the \$29-billion poultry industry of the United States.

This year, the US departments of agriculture and the interior will lead a \$29 million effort to test wild birds for H5N1 and other avian flu viruses in Alaska and other parts of the United States. “We’re at the nexus of bird migration from the Canadian Arctic to Russia and southeast Asia,” says Robert Leedy, chief of the US Fish and Wildlife Service’s office of migratory bird management in Anchorage, Alaska. “If H5N1 is going to be transferred in wild birds, the most likely avenue is Alaska.”

Crossing continents

But some scientists have reservations about the testing programme. Many flu experts think poultry smuggling or imports, rather than migrating birds, are far more likely to bring in the virus. And others point out that it’s still not clear how or whether wild birds contribute to H5N1 outbreaks in domestic poultry. Given these uncertainties, some question the decision to spend millions of dollars hunting for flu in Alaska, when the H5N1 virus is already racing across the rest of the globe. “More information is always better, so you can’t complain about that,” says William Karesh, director of the Wildlife Conservation Society’s Field Veterinary Program based in New York City. “But it’s much more important to go where the disease is in the developing countries, to see how this thing is spreading.”

Until last year, no one thought that migratory birds played any serious role in the spread of H5N1. But in July 2005, a team of virologists reported that some 6,000 migratory birds had died of an H5N1 outbreak at the Qinghai Lake nature reserve in China¹. Many of the dead birds were bar-headed geese, which fly from China to India and Myanmar every year. Since that report, the H5N1 strain has been found in dead migratory birds in Asia, Russia, Europe, Africa and the Middle East. Four people even died from bird flu after collecting feathers from infected wild swans in Azerbaijan². So what role do migratory birds play in spreading H5N1 around the world?

Genetic studies may help to answer this question. This May, Ian Brown of the United Kingdom’s Veterinary Laboratories Agency in

Weybridge revealed that H5N1 viruses taken from dead wild birds in Europe are very similar to H5N1 viruses found in Mongolia, Siberia and Qinghai Lake. Scientists have also reported that healthy birds in China were carrying the H5N1 strain just before their autumn migration last year³. That suggests the birds could have caused the outbreak of the virus in Europe last autumn, by carrying it to the continent from east Asia.

Other studies have implied that wild birds shuttled the virus between Europe and Africa, where H5N1 first showed up in February. A team of researchers recently suggested that

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— William Karesh

migrating birds may have transmitted H5N1 to Nigeria, the first African country to report the virus⁴. The scientists sequenced genes from bird flu viruses found in chickens on poultry farms. They discovered that many of the viruses, which seemed to cluster into three genetic groups, were similar to those found on other continents, including one strain that has been found only in wild birds in Europe. What’s more, the virus outbreaks in poultry were found along major bird migration corridors.

But none of these studies can conclusively show that migratory birds transmit the virus. In all cases, the wild birds themselves could have caught H5N1 from poultry or from some ‘bridge’ group, such as crows, jays or grackles. Officials agree that answering questions about the role of wild birds will require a lot more field work in live, migrating birds.

And that’s why James Sedinger and his team of young biologists, including Comstock, are spending their summer swabbing birds’ rears on the Yukon Delta.

Sedinger, a wildlife biologist at the University of Nevada at Reno, has been studying migratory birds in Alaska since 1977. His field camp sits at the edge of the tidal Tutakoke River, near where it runs into the Bering Sea. The camp rattles with the noise of birds at all hours in summer, when the sun sets for three hours every night, and if you don’t watch your step you’re likely to stumble into a mother bird sitting on a nest tucked into the grass.

This is breeding central, not just for migrating eiders, but also for black brants — chunky sea geese that fly from Alaska to points south for the winter. Over decades of work, Sedinger has snapped identifying bands on thousands of brants at Tutakoke. Some of the birds have been found to spend the winter in China, Japan and Korea. The US government’s plan to head off bird flu focuses on about 29 such species that spend time in Alaska and travel to Asia. About 15,000 birds will be tested in the state. Most will be trapped live by biologists. But the Department of Interior will also test birds shot by sport and subsistence hunters and — if and when they happen — dead ones found among mass bird die-offs.

Out for the count

So far, 3,772 samples from Alaska have been tested, and none has turned up positive. But that’s not surprising. Most of the samples taken from live wild birds around the world are clear of H5N1, says Ward Hagemeyer of Wetlands International, a group that coordinates volunteer surveys of migratory birds. Last winter, Wetlands International tested almost 6,000 wild birds for H5N1 along migration paths in Africa, Europe and Asia. And scientists

funded by the European Union have tested a total of 45,000 wild birds in Europe since last autumn. So far, none has turned up positive for H5N1, even though dead wild birds in all of these places have been found to be carrying the virus. Migrating birds are known to carry avian influenza strains other than H5N1. But, says Hagemeyer, "Finding H5N1 in healthy wild birds is amazingly difficult."

That perplexes wildlife-health specialists. Many of the dead or dying migratory birds found to be carrying H5N1 have been found near poultry farms, so it's not clear whether the wild birds infected the poultry or vice versa. And nobody knows how long the birds survive with H5N1, whether they are able to transmit the virus to other species and, if so, for how long they are contagious.

Hagemeyer cautions that even the most incriminating data pointing to the role of wild birds in transmitting H5N1 — the genetic study of H5N1 in Nigeria — leave some gaps. Nigeria doesn't conduct rigorous safety checks on imported poultry. So it's possible that shipments of infected birds seeded the outbreak, a point the original research team concedes. The finding that one of the Nigerian strains matches a strain found only in wild European birds also isn't convincing evidence that migratory birds were the cause of the outbreak, says Hagemeyer. This is because many more H5N1 strains have been studied from wild birds than from poultry. "If you're looking at relations between strains, you're far more likely to find a close relative in the wild bird database than in the database for poultry, because more of them have been sequenced," he says.

Needle in a haystack

Hagemeyer adds that wild birds "probably play a role, but we still haven't found the smoking gun". Other animal-health officials agree. The real question is whether wild birds can serve as a permanent reservoir for the virus, rather than simply transport it from place to place.

That is a worrying possibility because whenever the virus has a chance to mix in large groups of animals, as in the huge poultry farms of Asia, it might mutate so that it becomes lethal to people. "There are slight differences between the strains we find in wild birds, which suggests something is happening," says Scott Newman, a veterinary health specialist with the Wildlife Conservation Society who works at the headquarters of the United Nations Food and Agriculture Organization, in Rome. "But it's hard to tell whether it's happening in wild birds or in poultry."

Given all these unknowns, some are sceptical about the United States investing so much money hunting for H5N1 in live birds. Newman points out that H5N1 has swept from Asia towards Europe, and not the other way around. Perhaps, he suggests, officials should be more concerned about migration from Europe, through Greenland to the US east coast, where limited testing is ongoing. Even the biologists



Biologists are taking faecal swabs off eggs and birds' rears in the Yukon Delta in Alaska to test for H5N1.

doing the work say it's unlikely that they will intercept the virus. "We're looking for a needle in a haystack," Sedinger says.

Others are frustrated that the United States is spending so much on testing birds within its own borders while the disease continues to spread elsewhere. "We should be doing more overseas," Karesh says. "I'd like to see the United States do more outreach."

The Food and Agriculture Organization is already working with groups such as Wetlands International, the Pasteur Institute, the French Agricultural Research Centre for International Development and the Centers for Disease Control and Prevention to test birds worldwide. This January, the United States pledged to spend \$334 million in international aid for

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countries battling influenza. And last month, the US Agency for International Development agreed to provide \$5 million to an effort called the Global Avian Influenza Network for Surveillance. Headed by the Wildlife Conservation Society, this will test birds for H5N1 in remote and poor places, such as Mongolia, that don't have their own surveillance system in place. This is the only good way to get a better handle on the dynamics of the virus in wild birds, Karesh says.

Meanwhile, biologists who normally struggle for grants to study the basic biology of migrating birds are frustrated by the huge

amounts of cash flowing for bird-flu studies. The field camp at Tutakoke, for instance, wouldn't even have been funded this summer without the bird-flu surveillance money.

Sedinger and other bird biologists say the paucity of long-term studies on migratory birds makes it difficult to understand the spread of H5N1. It wasn't until 1995, for example, that scientists discovered that spectacled eiders spend their winters floating in holes in pack ice in the Bering Sea⁵.

So while biologists are thankful for the bird-flu surveillance money, they also wonder whether officials will take the logical next step and invest more in monitoring studies as well.

Still, biologists are happy for any chance to learn more about migratory birds — especially if it means spending long days chasing after ducks and geese on wind-blown tundra. And even if they don't track down the virus in Alaska, Sedinger, Comstock and the rest of the Tutakoke team will learn more about how viruses circulate in and between species — an important area of research given that at least two human flu pandemics in the past century began as bird viruses. "We're going to get our hands on some birds we wouldn't normally get to study," says Leedy. "That's going to lead to some good work."

Erika Check covers the biomedical sciences for Nature.

1. Chen, H. *et al. Nature* **436**, 191-192 (2005).
2. Eurosurveillance www.eurosurveillance.org/em/v11n05/1105-222.asp (2006).
3. Chen, H. *et al. Proc. Natl Acad. Sci. USA* **103**, 2845-2850 (2006).
4. Ducatez, M. F. *et al. Nature* **442**, 37 (2006).
5. Petersen, M. R., Larned, W. W. & Douglas, D. C. *Auk* **116**, 1009-1020 (1999).

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