

On 19 March, a private Learjet touched down at the Munich airport. Inside the flying ambulance was a 73-year-old man from Abu Dhabi suffering from severe pneumonia. His family hoped German physicians might save his life.

They couldn't. A week later, the patient died at the Schwabing Clinic in northern Munich; his body was returned to the United Arab Emirates the next day. By then lab tests had shown that he was infected with a new coronavirus—the 17th known case worldwide and the 11th fatal victim. German officials have not revealed the man's identity, although that didn't stop the German tabloid *Bild* from claiming that he was “a sheik” from a “ruling family.”

Whoever he was, the man's final journey provided scientists another opportunity to learn something about a respiratory disease outbreak that is still surrounded by questions. As *Science* went to press this week, University of Bonn virologist Christian Drosten said he had already sequenced the genome of the virus and would soon publish a comparison with other known sequences that may shed some light on the pattern of spread of nCoV (for novel coronavirus), as the World Health Organization (WHO) has dubbed the pathogen.

Six months after the virus was first reported, scientists are convinced that there

are more people infected than the 17 known cases, some perhaps with cases so mild that they aren't seeking care, but they don't know how many; they assume the virus originates in animals, but they don't know in which species. There is almost certainly human-to-human transmission, but it's unclear how efficiently it occurs. The virus is a distant relative of SARS, the disease that terrified the world in 2003, but it's unknown if nCoV, too, has the potential to explode into a global health crisis.

Part of the problem is that the affected countries have released little information about the outbreak so far and appear reluctant to collaborate with foreign researchers eager to find out more (*Science*, 15 March, p. 1264). Saudi Arabia, for instance, has reported nine cases to WHO, but for some, there is no information about patient age, sex, place of residence, or circumstances surrounding the infection. “A lot of key epidemiological information is missing,” says virologist Vincent Munster of the Rocky Mountain Laboratories in Hamilton, Montana, which is part of the National Institute of Allergy and Infectious Diseases.

Since the virus emerged, scientists have published genomes of four strains isolated from patients, developed diagnostic tests, and set out to develop animal models—with the first successful attempt published online

this week by Munster's team in *The New England Journal of Medicine* (*NEJM*). In a paper published in the 14 March issue of *Nature*, a team led by Bart Haagmans of Erasmus MC in Rotterdam, the Netherlands, also identified the virus's receptor, a well-known protein called DPP4 that sits on the surface of cells deep inside the human lung.

So far, the virus appears to circulate primarily in the Arabian Peninsula. Beside the nine patients from Saudi Arabia, there were two from Qatar, and the first two confirmed cases are now known to have occurred almost a year ago during an outbreak in Jordan. (They weren't identified and reported until last fall.) Three U.K. residents were also infected—two fatally—in January and February, but the first one of those had traveled to Saudi Arabia and Pakistan before falling ill and presumably picked up the virus on the way. That two of his family members became infected suggests that the virus spreads from human to human—although apparently not very efficiently, because 135 of the patients' contacts weren't infected.

Munster's short paper published online in *NEJM* this week not only provides researchers with a way to test candidate drugs and vaccines, but also offers formal proof that nCoV—often called hCoV-EMC, a name given to it by Erasmus MC researchers who first sequenced it—is the cause of the human disease. For the study, Munster and his colleagues inoculated six rhesus macaques with the virus. They developed pneumonia within 24 hours, with symptoms such as fever, cough, and reduced appetite, and necropsy revealed bright red lesions throughout their lower respiratory tracts. (The team's pathologist called the severity of the disease—which varied from one animal to the next—“mild to moderate.”) The researchers reisolated the virus from the sick animals, thus fulfilling Koch's postulates, the classic set of criteria used to prove that a pathogen causes a disease.

Haagmans's group at Erasmus MC has tried the same experiment in cynomolgus macaques, but in that species the virus replicated poorly and didn't cause disease symptoms at all. Haagmans presented those findings at a Rotterdam meeting on 14 March, but says he wants to better understand why the animals were unaffected before publishing the data. It could be something species-specific, he says, or perhaps the monkeys were too young; previous work by the same group showed that older monkeys were much more susceptible to the SARS virus, a pattern that jibed with the

human outbreak. (Munster's rhesus monkeys were between 6 and 12 years old.)

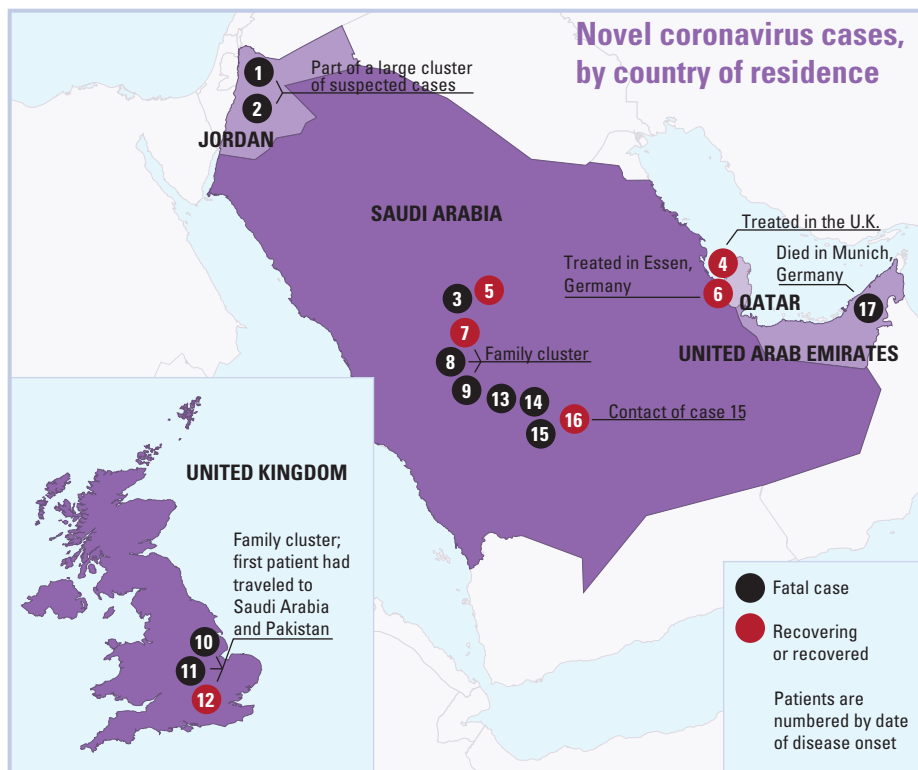
But few labs have monkeys, and they're expensive to work with; it's "really critical" that researchers also develop a small-animal model of infection and disease, says Matthew Frieman, a coronavirus researcher at the University of Maryland School of Medicine in Baltimore. Although the SARS virus does replicate in mice, and a mouse-adapted version makes them sick as well, Frieman and others have been unable to infect mice with the new virus. Munster has tried in vain with hamsters, and Haagmans's attempt in ferrets has failed as well. "We seem to have little luck with small animals," Haagmans says.

There are other ways of gauging the pathogenic potential of the new virus. A group led by Kwok-Yung Yuen at the University of Hong Kong tested whether it could infect a total of 27 cell lines from humans and animals, as a guide to which tissues and organs the virus might infect in the real world. In a paper published online by the *Journal of Infectious Diseases* on 26 March, they reported that nCoV replicated in 16 of the cell lines, more than any other known coronavirus—including human lung, kidney, intestinal, and liver cells. That might help explain why known cases of the disease have been so severe, the authors said.

But other researchers balked at that. In an accompanying commentary, Kenneth McIntosh of Boston Children's Hospital cautioned that studies of human cell lines—including some derived from tumors—often have little power to predict what happens in patients. "I think they drew too many conclusions," Frieman adds. In an e-mail to *Science*, Yuen says his group is well aware of the study's limitations, but writes that absent an animal model, the cell line approach "is all we can do at this moment."

Given the many questions about the epidemiology in the Arabian Peninsula, some researchers are hoping that the genomes of viruses found at different times and locales may yield clues about what is happening, an approach known as genomic epidemiology. So far, viral genomes from four patients have been published: the first reported case, a man who died in Jeddah in June and whose virus was sent to Erasmus MC; a patient who traveled to the United Kingdom in September for treatment; the first case in the U.K. family cluster earlier this year; and one of the Jordan patients, whose virus was obtained and sequenced by a U.S. Navy laboratory in Cairo.

In a recent paper in *Emerging Infectious Diseases*, a U.K. team published its genomic analysis of the first two cases; team member Andrew Rambaut, an evolutionary biologist



of the University of Edinburgh, then updated the analysis on his own Web site after two more genomes became available. The team found considerable variation between the four genomes, and calculations suggest that the virus has circulated since 2011, Rambaut says. In theory, it may have done so only in humans—assuming that many infections were mild and went undetected. But given today's travel patterns, it's unlikely that the virus would remain confined to the Arabian Peninsula if that were the case, Rambaut says. A more likely scenario is that it's circulating in animals and was repeatedly introduced into the human population, he adds—perhaps setting off short human-to-human chains of transmission now and then.

Drosten says that he soon plans to publish his own analysis, which will also include the newly sequenced virus from the Munich patient. In addition, Drosten says that he has an unpublished, partial sequence from a Qatari man who was treated at a hospital in Essen, Germany, last fall. (His group had huge trouble sequencing the genome because the samples contained very little viral RNA.) Drosten's analysis shows that the viruses from Qatar and the United Arab Emirates, which are geographically close, form a separate group—Drosten calls it the "Gulf clade"—from those in Saudi Arabia and Jordan. But he declined to speculate what, if anything, this says about the epidemiology.

Which animal species serves as the springboard to humans isn't clear, and there could be more than one. The viral sequences found so

far are closely related to bat coronaviruses, but bat-to-human virus jumps are rare because contact between the two species is uncommon. One notable exception is the Nipah virus, which is believed to cause human disease occasionally in Bangladesh when urine from fruit bats contaminates date palm juice. The alternative is that there is some intermediate animal host, Munster says.

Some anecdotal reports have suggested that livestock acted as that bridge species. German media have reported Clemens Wendtner, a physician at the Schwabing Clinic, as saying that the Abu Dhabi patient owned racing camels—a popular pastime among the Gulf's elite—and that he may have become infected when he visited them shortly before falling ill. The Qatari patient treated in Essen owned a camel and goat farm and told his German doctors that some of his goats had been sick before he fell ill himself; one of his animal caretakers also had a severe cough and was hospitalized.

Only epidemiology studies on the ground can bring more light to many of the questions, Munster says. Whether they are taking place in the four affected Middle Eastern countries is unclear even to WHO, which is trying to stimulate a concerted research effort. "I don't know if WHO has a full handle on all of the studies that are being done or are in the works," WHO spokesperson Gregory Hartl says—and as a human health agency, WHO is not involved in animal studies, he points out. "But we do need to know more about this virus." —MARTIN ENSERINK

## PHENOMENA: THE LOOM

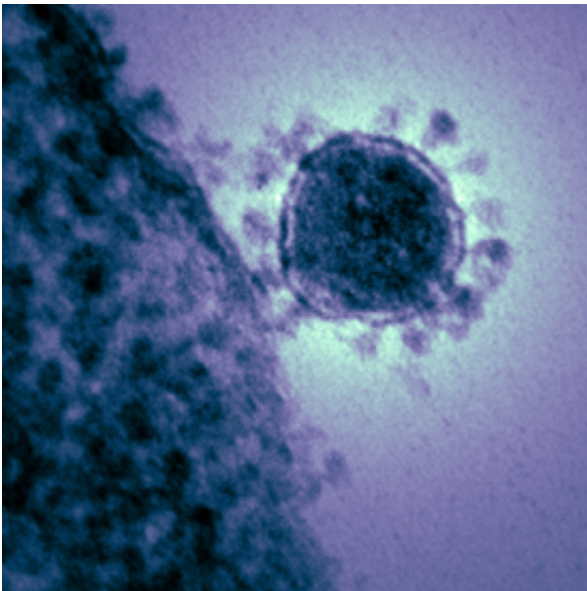


Photo by Bruce Dale/National Geographic

## THE LOOM:

# Listen Closely To The Bats and You Can Hear the Viral Chatter

by Carl Zimmer



HCoV-EMC, by Beth Fischer. Source: NIAID/NIH

Last June, a sixty-year-old man in Saudi Arabia fell ill with pneumonia. His disease, [it turned out](#), was caused by a virus no one had seen before. It was a coronavirus—in other words, it belonged to a lineage of viruses that includes ones that cause colds as well as ones that cause [SARS](#). But this new virus was genetically distinct enough to be considered a species in its own right. Scientists now refer to it by the dreary, unpronounceable abbreviation HCoV-EMC. Eleven days after being admitted to a Jeddah hospital, the man infected with this new

virus died.

A single death from a new virus is hardly unheard of. But over the past few months, virus-watchers have gotten increasingly anxious about HCoV-EMC. [So far](#), 15 people have been diagnosed with the virus, and nine have died. While some victims have turned up as far away as England, everyone with HCoV-EMC has had some connection to the Arabian

Peninsula. Some victims belonged to the same family, suggesting that the virus can spread from one person to the next.

We can't say for sure whether we're at the beginning of a HCoV-EMC pandemic, or at the end of a minor outbreak, or experiencing something in between. But scientists are not waiting around until the virus has finished traveling down whatever path it will take. They're working hard to figure out the biology of the virus, and they're also trying to figure out its history. How it got into 15 people over the past 9 months might give us a hint as to what it may do in the future.

In other words, scientists have to probe the evolution of HCoV-EMC.

The closest relatives to HCoV-EMC are coronaviruses that live in European bats. That doesn't mean HCoV-EMC came from Europe, however. Most of the diversity of coronaviruses is unmapped, especially the ones that live in animals. There could well be bats in the Near East with more closely related but undocumented coronaviruses. The fact that all the human victims were at some point in the Arabian Peninsula certainly raises the possibility that bats there spread the virus to people.

The link between bats, coronaviruses, and humans is a familiar one. SARS moved from bats to humans ten years ago, and since then scientists have found other coronaviruses that moved from bats to humans. The biology of HCoV-EMC itself offers more evidence for how this might have happened. Some viruses are very fussy about how they invade cells. They only infect one type of cell in one species. But HCoV-EMC is a lot less picky. It latches onto a receptor on cells lining the airway, and scientists have found that it can invade airway cells from not just humans, but pigs and bats. The ancestors of HCoV-EMC might have lived in bats and yet they may have already been prepared to infect humans.

Recently, a team of evolutionary biologists began to draw an evolutionary tree of HCoV-EMC based on a comparison of the viruses isolated from three victims—the first patient from June, and two later victims from England. They identified mutations that arose in each of the three lineages of viruses since they diverged from a common ancestor. Mutations accumulate at a roughly clock-like rate, which means that scientists can use them to estimate how long ago lineages split apart. In the case of the three HCoV-EMC viruses, their common ancestor dates back to 2009.

Three years is a long time for a virus to be circulating among people without anyone noticing. It's conceivable that a lot of people passed it around and only got mildly sick.



But the fact that nine out of fifteen people identified so far with HCoV-EMC actually died suggests that this is a fairly deadly virus, making invisibility unlikely.

If that explanation fails, what happened? Virologists I've spoken to favor a phenomenon known as ["viral chatter."](#) Viruses don't just barge across the species barrier in one great rush. They tentatively make incursions—many of them spread across years. The SARS virus, for example, infected a few people before becoming a massive epidemic in 2003. During these incursions, animal viruses may acquire mutations that gradually prepare them to become good at spreading from person to person.

For now, thankfully, HCoV-EMC seems to be [bad at that kind of transmission](#). Each sick person made contact with many others and almost none of the exposed people got sick as a result. Instead, people must be getting infected through contact with sick animals. They're picking up viruses that diverged from a common ancestor a few years ago and are still circulating among animals.

It's not likely that humans are getting sick from HCoV-EMC thanks to frequent contact with bats. When's the last time you gave a bat a kiss? Instead, HCoV-EMC may be using a stepping-stone species to get from bats to humans.

Again, SARS offers some lessons. It appears that the SARS virus spread from Chinese horseshoe bats to civets, cat-like animals that live in East Asia, and then to humans.

A decade after the SARS epidemic, no one can say for sure how SARS got from bats to civets. But the answer must lie somewhere in their ecology. Matt Frieman, a virologist at the University of Maryland who studies bat coronaviruses, pointed me to an example of the interactions between bats and civets buried in [a 2010 article about civets and coffee](#).

Civet cats eat wild coffee cherries and then poop them out. People collect their poop and clean off the beans they contain. The resulting coffee, the article claims, has a taste that's "smooth, chocolaty and devoid of any bitter aftertaste." Western appetite for this delicious coffee has led people to hunt for civet poop in forests, and to feed captive civets coffee beans on little farms.

One passage in the story should give you a jolt. A coffee supplier inspecting a batch of civet-processed beans knocks down his buying price because of some impurities: "inferior beans that the civet had spat out; beans chewed on, not by civets, but bats."

So imagine SARS-infected bats in a rain forest chewing on the same coffee cherries as civets. It's enough to start an epidemic, perhaps.

Ten years later, a continent away, bats with HCoV-EMC may be coming into contact with other animals as well. Goats drinking at watering holes might spend time near bats in neighboring fruit trees, for example. And the farmers of the goats might then pick up viruses from their livestock. At this point, it's a notion—or, rather, a scientific hypothesis. The animals of the Near East will tell us whether it's a good one or not.

(For more on the emergence of new viruses, see my book [\*A Planet of Viruses\*](#).)