NEWS FEATURE

News Feature: Many species, one health

In the race to save endangered frogs from extinction, disease ecologists are hunting for patterns of infection that could also improve human well-being.

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Frogs infected with the flatworm *Ribeiroia* could be extras in a horror movie. Their bodies are a tangled mess, sprouting extra limbs and joints, and covered with strange skin webbings and bony protrusions. No longer able to walk or swim, they have become easy prey for hungry birds and that's exactly what the parasite is counting on.

Ribeiroia is a trematode, a type of flatworm that lives in several hosts, jumping between frogs, birds, and snails. Once a bird eats an infected frog, the worms reproduce sexually within the bird's body and their eggs hitch a ride on the bird's feces, making their way into ponds and marshes. When they hatch, the eggs release larvae that infect snails. Once inside, the larvae reproduce asexually to produce another form of larva that creates cysts around the developing limbs of tadpoles. The result is not only visually ghastly, but a harbinger of a wider problem across multiple animal populations.

Work on *Ribeiroia*'s exploits is one front in a wider campaign by a small but growing group of researchers. Disease ecologists are closely tracking and modeling disease systems in an ambitious bid to determine the underlying principles of how infections spread. The work could eventually help land managers to reduce disease impacts by making a few ecosystem tweaks, perhaps reintroducing a species in one location or repelling another species somewhere else. However, by uncovering the relationships between multiple pathogens and multiple hosts, researchers are also revealing connections between human and animal health. In the past decade, the concept of "One Health," first used in the 1800s to suggest crucial links between the health of animal and human populations, has gained traction among public health and disease researchers: the Centers for Disease Control and Prevention (CDC) established a One Health office in 2009, and a year later the CDC, World Health Organization, and others devised steps to adopt a One Health approach that mingles research in animal and human

Researchers from the University of Colorado and the City of Gresham, Oregon, search for malformed amphibians in a pond near Portland, Oregon. Image courtesy of Dave Herasimtschuk © Freshwaters Illustrated.

The parasitic flatworm *Ribeira* causes limb deformities in Northern leopard frogs such as this one. Image courtesy of Dave Herasimtschuk © Freshwaters Illustrated.

disease in hopes of stemming the spread of pathogens among species. Proponents say that understanding disease spread among wildlife could lead to a new approach to fighting human infection: using ecological management to prevent, predict, or control outbreaks. Critics, though, argue that such an approach is at best an oversimplification.

Whatever the case, new approaches will be essential if future outbreaks are going to be stamped out efficiently. "We're being confronted with emerging infectious diseases all the time, and at an increasing rate," says Rick Ostfeld, senior scientist at the Cary Institute of Ecosystem Studies in Millbrook, New York. "The response of our public health community is vigorous, aggressive, and high-tech, laudable in many ways, but inadequate." Ostfeld says that, even after an outbreak has fizzled, infectious disease specialists often know little about what caused the outbreak or how they might be able to react more quickly the next time.

Battling outbreaks after they've begun, says Ostfeld, is like pouring water on a raging inferno. Instead, he envisions public health officials behaving more like fire marshals, detecting when conditions are ripe for disaster and acting to reduce the risk of an outbreak. "If some large development project is going to reduce protective diversity, or cause human encroachment into dangerous habitats, or cause vector populations to explode, we might be able to alter the plan to avoid the disease outbreak," suggests Ostfeld. That plan might involve protecting a habitat for species that inhibit the spread of disease, for example. Alternatively, if ecological insights enabled health officials to anticipate a likely outbreak, they could deploy vaccines much more quickly.

A lot more work will be needed before such approaches are ready for routine use by health agencies like the CDC. The interactions between hosts, pathogens, and environmental factors are complex and sometimes counterintuitive. Until recently, however, disease ecologists have focused mainly on simple systems involving one host and one pathogen. These systems are relatively easy to model and understand, but they are not very realistic or useful. Researchers will have to grapple with the fact that, as in the case of *Ribeiroia*, individual hosts often harbor multiple pathogens, Ostfeld says. Figuring out how all these interactions affect a pathogen's virulence is the ultimate goal, he says. "That's our challenge over the next 5 to 10 years."

Pond Life

Sightings of malformed amphibians across the Western United States have skyrocketed since the mid-1990s. In some ponds, the level of infection is so severe that biologists are worried the disease could wipe out certain species. "We visit ponds where 70–80% of the endangered species, like the California red-legged frog, are so deformed they're likely to die," says Pieter Johnson, a disease ecologist at the University of Colorado, Boulder, who has studied this disease system for decades.

Johnson began studying frog deformations as an undergraduate student at Stanford University, California, in the late 1990s. Landowners in Santa Clara County had begun reporting severely malformed Pacific treefrogs (*Pseudacris regilla*) in their ponds, and Johnson noticed that many other species were suffering the same fate, including American bullfrogs (*Rana catesbeiana*), Western toads (*Bufo boreas*), and California newts (*Taricha torosa*). However, the only ponds affected were those that hosted a particular group of snails, a group that is the exclusive first intermediate host of the parasite *Ribeiroia ondatrae*.

Scientists had already raised the possibility that parasites could be the cause of the amphibian malformations based on dissections of the dead and diseased animals. But Johnson and other Stanford biologists were the first to test the theory in a laboratory by exposing tadpoles to *Ribeiroia* larvae. Extending their search across the Western states, they found nearly a dozen species of frogs, toads, and salamanders were vulnerable to *Ribeiroia*-induced deformities. Infection hotspots, they noticed, tended to be most common along major bird flyways (1–3).

Although long-term data are hard to come by, Johnson believes the rate of malformations caused by *Ribeiroia* infections has risen. A review of old field notes, historical surveys, and dissections of old museum specimens seem to indicate that malformations used to be less common. The change, he says, is probably because of a host of factors: the increasing eutrophication of waterways, which delivers an oversupply of nutrients that boosts snail populations; growing use of pesticides that reduce amphibian immunity; changes in biodiversity; and shifts in bird population dynamics. "Most of the hotspot sites are ponds greatly modified by human activity," says Johnson. "So differentiating among these factors is a bit challenging."

It was unclear, for example, whether disease-transmission rates were greater in ponds with lower or higher biodiversity. On the one hand, adding extra host species to an ecosystem generally reduces the overall rates of disease, a phenomenon known as the dilution effect. However, at the same time, a broader range of host species will generally support a wider variety of parasites, raising the chances of infection (4). Each factor pulls in the opposite direction, creating complex scenarios that vary depending on the ecosystem and the species.

To help solve the puzzle, Johnson and his team ventured into hundreds of San Francisco Bay Area ponds, collecting amphibians and sending them to Colorado for dissection. "It was pretty mucky work at times," says doctoral student Daniel Preston. "Some of the

trematode parasites we work with can accidentally swim into you and cause a nasty rash, so we always wore waders. These aren't ponds you'd want your kids playing in."

The researchers also harvested live parasites from snails—often in the middle of the night, when the parasites were most active—and introduced them into four different types of outdoor experimental enclosures. One had lots of parasite species, a second had lots of hosts, a third had plenty of both, and a fourth had few of either. Then the researchers dissected the hosts to count how many parasites had successfully made the jump.

Their quantitative approach was designed to disentangle the relative contributions of host and parasite diversity. Introducing more host species, for example, corresponded to a dramatic fall of up to 65% in *Ribeiroia* infections. In the laboratory, communities with three species of hosts saw about 40% fewer overall parasite infections than communities with just one species. A community relatively abundant in parasite or pathogen species also saw a drop in the amount of disease transmission, although the effect was not as great (5).

Parasite competition may be the key factor behind this observation, Johnson believes. As parasites begin to infect a host, the hosts' immune systems ramp up, making it more difficult for other types of parasite to join the party. That finding suggests it may be possible to control the disease by managing the biodiversity in ponds, identifying and promoting the key species that keep *Ribeiroia* in check. Similar strategies could be applied more broadly to address the global decline in amphibian populations.

Beasts in the Field

Ostfeld is studying a different ecosystem to investigate why some species are so good at spreading disease. The best ones, he suggests "live fast and die young," and reproduce explosively. In a recent study he suggests that this abundance of hosts means that a pathogen is more likely to encounter them, and tends to be well adapted for infection perfection. These hosts also may have poor immune systems, rendering them unable to resist infection (6).

Ostfeld's specialty is tick-borne Lyme disease, a potentially debilitating bacterial infection spread by ticks to humans, which is the most commonly reported vector-borne illness in the United States. In 2012, the CDC recorded more than 30,000 probable cases of Lyme. The primary tick host is the white-footed mouse, says Ostfeld. "That's the guy who basically never disappears, who's present in all disease systems, from the most species-poor to the most species-rich," he notes. Other host species tend to reduce overall tick numbers. Possums and gray squirrels, for example, gobble up ticks as they groom themselves. "There are actually some interesting parallels with Pieter's work," says Ostfeld. He believes that promoting rich, diverse ecosystems that include long-lived animals should help to protect wildlife and humans against disease (7). Keystone disease reservoirs, he says, should be monitored and managed.

However, this suggestion—and the hunt for rules that apply to multiple ecosystems—has been met with skepticism in some circles of disease ecology. Retired professor of parasite ecology Sarah Randolph, formerly at the University of Oxford in the United Kingdom, has argued that the dilution effect is an overly simple and optimistic idea. Protecting biodiversity is intrinsically valuable, she argues, leaving no need to make the case for dubious utilitarian goals, such as protecting human health (4). Other researchers have said that although conservation might improve public health initiatives in some cases, the general link between biodiversity and disease is weak and idiosyncratic. Disease risk is more likely a product of a host and vector's particular biology, and not based on patterns of species biodiversity, according to Dan Salkeld, a research associate at the Stanford Woods Institute for the Environment in California (8).

Ostfeld acknowledges that it is not always possible to draw general rules from studies of ecosystem interactions. Invasive exotic species, for example, do not always displace natives and damage their ecosystems. "But the fact that there are exceptions shouldn't prevent us from applying policies that reduce invasive species," he says.

Health for All

Johnson's and Ostfeld's work plays into the broader "one health" idea within disease ecology. The CDC's recently opened One Health office helps veterinarians and public health experts work with agencies like the US Department of Agriculture to control zoonotic diseases. The CDC has also brought an ecology team into their viral pathogens team to help respond to the current Ebola outbreak. "I really do buy into the idea," says Sonia Altizer, a disease ecologist at the University of Georgia, Athens, who studies how human changes affect the spread of pathogens in wildlife. Humans are putting such great demands on the planet that new diseases are bound to emerge, she says.

Humans' connection with other species is clearly a two-way street. Although we often think of diseases jumping from wildlife to humans, the leap can happen in both directions, says Altizer. Outbreaks of measles and other human-spread respiratory infections, for example, have caused disease outbreaks and deaths among endangered chimpanzees in Uganda. As their habitat becomes increasingly fragmented, chimpanzees overlap more with humans, making it more likely that they will exchange illnesses. "These apes are not in a natural situation anymore," says Julie Rushmore, one of Altizer's former graduate students, who has studied chimpanzees' social networks in Kibale National Park, Uganda. "A lot of the pathogens they're exposed to are coming from humans, either through tourism, research, or living in an overlapping landscape with humans," she says.

Rushmore is now developing strategies for administering vaccines to these endangered animals when diseases strike. Her studies, based on human epidemiology, have helped determine which individuals are most important to target (9). Some apes, especially high-ranking mothers, form day-care groups to share child rearing, and these apes can become nodes of infection, just as certain humans have been known to act as "superspreaders."

Patterns like these may not be universal, but Ostfeld believes that they are worth searching for. "Maybe it's possible that everything is idiosyncratic, maybe there are no principles," he admits, "but, on the chance that there are, it's worth pursuing them."

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