Courtesy Brian Langerhans

Catching
Evolution in the Act

CALS biologist explores predictability of evolution in Bahamian blue holes.

By Dee Shore

For biologist Brian Langerhans, cerulean sinkholes that dot the Bahamas are places of mystery and intrigue – environments that contain clues to help answer one of science's most captivating questions: How predictable, he wants to know, is the course of evolution?

Dr. Langerhans is an assistant professor in the College of Agriculture and Life Sciences' Department of Biology, and for the past decade he has been studying fish of the genus *Gambusia* living in submerged Bahamian caves known as blue holes.

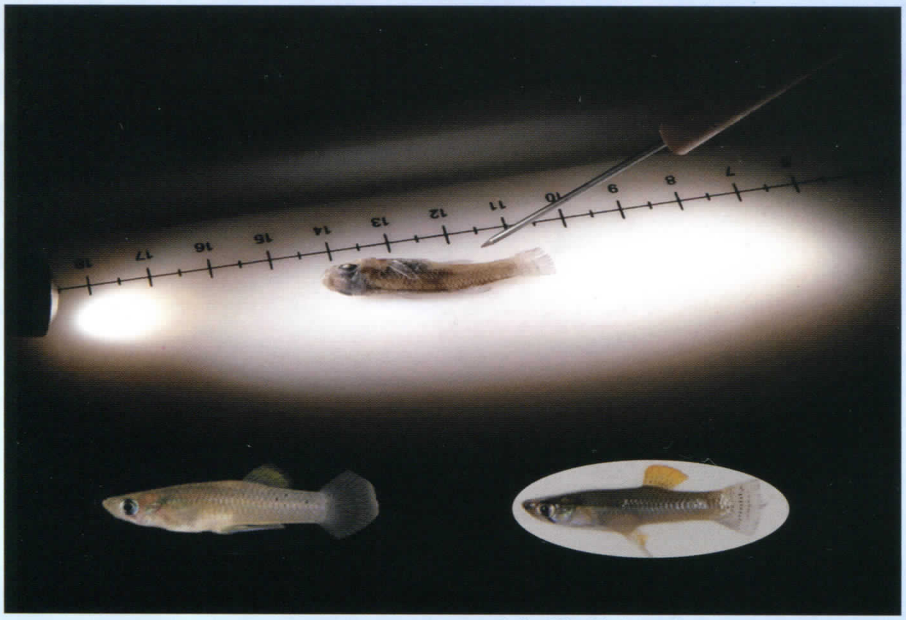
The deep, water-filled holes were dry during the last Ice Age, but as sea levels began to rise about 15,000 years ago, the water table rose and flooded the ancient caves from the bottom up. Soon, fish colonized many of these newly aquatic environments, but different kinds of fish wound up in different blue holes.

Most of the blue holes – at least on Andros Island, where Langerhans conducts much of his research – have *Gambusia.* And some, but not all, of those holes have bigmouth sleepers, a larger species of fish that preys on the *Gambusia.*

"The inland blue holes are really isolated environments – basically aquatic islands in a sea of land," Langerhans says. Paired with *Gambusia*, a diverse genus of fish that bear live young, the blue holes become veritable test tubes for examining all sorts of questions about how the fish – and possibly other animals – have evolved.

"It's an interesting system for studying all kinds of evolutionary questions," Langerhans explains. "We focus on the predictability of evolution: In other words, if we understand enough about organisms, can we actually predict the course of evolutionary change across different environments? Can we identify what causes changes within species, and what drives the formation of new species? And then, what explains these broader patterns among taxa – the major macro-evolutionary patterns across species?"

Inland blue holes, submerged caves in the Bahamas, are the homes of the small fish called *Gambusia*. The isolated environments made an ideal site for studying evolutionary questions.



Becky Kirkland (background); Insets Courtesy Brian Langerhans

The varying sizes of the *Gambusia* heads and tails, affecting their speed, reflect the extent of predation in their environment.

In the past, some biologists argued that evolution depends on too many chance events to be predictable. But more recently, studies on organisms as diverse as bacteria, tiny worms, fruit flies – and now, *Gambusia* – suggest that evolution can be replicated. And if it can be replicated, some biologists believe, it can, at least to some degree, be predicted.

And that leads to the questions that occupy Langerhans. To get at the answers, he has been studying *Gambusia* since the early 2000s, when he was a graduate student at Texas A&M University. While only two species of *Gambusia* inhabit most of the United States, Texas harbored no less than nine native *Gambusia* species, and there are more than 40 known *Gambusia* species in the world.

What, Langerhans wondered, was behind that diversity? It was in 2002 when Langerhans saw his first blue holes and his ideas about the predictability of evolution began taking shape.

"It was evident right away – the fish were behaving differently in blue holes with and without predatory fish. They looked different. Just by walking up and looking, I thought, 'Something really interesting is happening here,'' he recalls.

Since then – through his Ph.D. studies at Harvard University, a post-doctoral fellowship at the University of Oklahoma and his appointment at N.C. State in 2010 – the scientist has immersed himself in the task of understanding everything he can about *Gambusia* living in the blue holes.

By observing the small, minnow-like fish in the wild and testing them in the lab, Langerhans and his lab colleagues – post-doctoral researchers as well as graduate and undergraduate students – have studied such traits as their body shape, how fast they can take off swimming, which mates they prefer, their coloration, their fat content, the number of babies that the females bear – the list goes on.

Based on general ideas of how fish swim and the advantages different types of swimming provide under different ecological contexts – specifically, with and without predators – Langerhans has created models to predict how traits might evolve, and he's tested them to see if those predictions held true in the blue holes.

And it turns out, Langerhans says, "there's a lot of predictable evolution in blue holes."

"Depending on predators being present or absent, we know what kind of body shape they are going to evolve. We know the general size of the male genitalia. We know how their locomotor abilities are going to evolve, their acceleration capacity, their endurance, many aspects of their life histories," he says.

"All these things are predicted by evolutionary theory, and it turns out that most of the predictions are met," he says. Although the measurements aren't precisely the same with every population, he adds, the fish do tend to evolve in the same direction, according to the level of predation in their environment.

To explain, he points to differences in body shape: When there are predators, Langerhans says, the fish's tail region tends to become larger, and their heads get smaller. That body shape lends itself to fast starts. "The fish go 'boom,'" he says, with a clap, "to get away from a predator."

But something very different happens to *Gambusia* that have evolved where there are no bigmouth sleepers. They tend to take on a streamlined shape.

"They are not fast any more, but they don't need to be that fast," he says. "Instead, they have high levels of endurance, so they can swim around all day long without using large amounts of energy."

That's important, he adds, because without predators, populations get denser, which means more mouths – and less food per mouth. "So it's all about cruising around the water finding food," Langerhans says. "These fish that are in the low-predation sites are always moving, looking for food. They've got to cruise around all the time."

The pattern Langerhans has seen – larger tails and smaller heads in high-predation sites, and more streamlined bodies in low-predation sites – occurs over and over, both in the blue holes and among generations of fish raised in Langerhans' labs.

"It's not," he says, "like there was a single evolutionary split, and then these fish moved around to different blue holes to find an environment that suits their phenotype. Molecular genetic evidence suggests that every time they get to a blue hole, these fish just evolve whatever body shape is needed in that blue hole."

The model also seems to work beyond the blue holes: Langerhans has analyzed other scientists' data related to body shape and predation and seen that his model holds with at least 20 fish species from six different families found in diverse environments around the world.

Natural selection – survival of the fittest, as Darwin put it – is at work, Langerhans says, and so is sexual selection. In low-predation environments, the females prefer the streamlined shape, and in high-predation environments, they like the opposite.

Even among different populations of the same species, *Gambusia* from high-predation environments tend not to interbreed with those from low-predation environments.

"These fish seem to hate each other. The body shape is wrong, the color is wrong, it's possible other things like behavioral and olfactory traits that we haven't examined yet are involved. Overall, all the evidence has shown that when given the opportunity they choose to spend much more time with fish from their own population – it's like the fish from the other population don't exist," he says.

"If you put them in the same tank, eventually they go to opposite sides of the tank and stay there. I've left them there 6 to 8 months, and they literally stay there," he adds. "It's like, 'I don't want to have anything to do with that guy.'"

Body shape, Langerhans believes, appears to be a clear factor in speciation, but it doesn't explain it completely. Right now, he's pursuing ideas related to differences in the size and structure of the male's sex organ, called the gonopodium, within species.

"It's pretty obvious if you evolve differences in the genitalia you can very quickly get reproductive isolation and form a new species," he says. "But so far, scientists haven't concentrated on within-species variation (in fish genitalia) – probably because it's usually presumed to be very small – but that's where you can catch evolution in action. As speciation is occurring, that's actually how you can figure out why it's evolving this way."

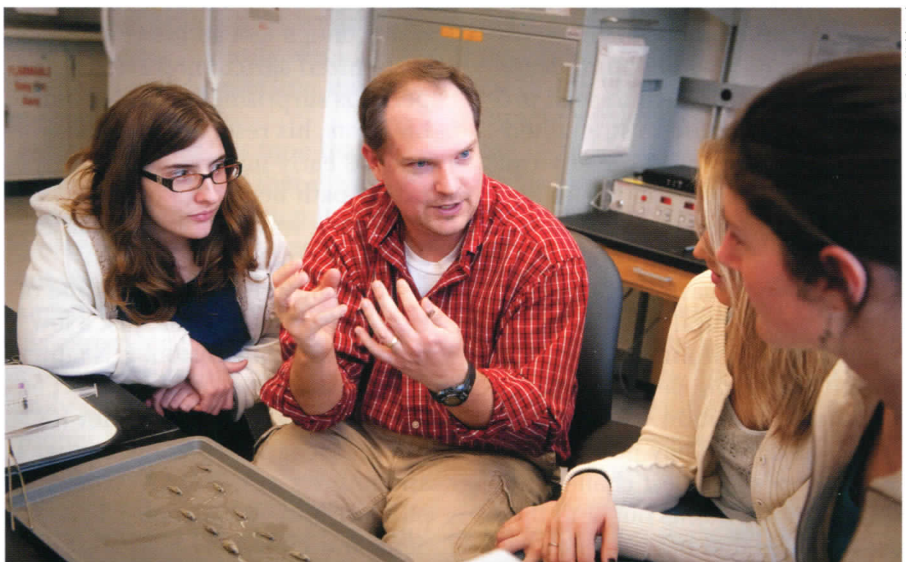
As Langerhans continues to study the predictability of evolution in the blue-hole *Gambusia*, he's also pursuing research aimed at understanding whether we can predict how the changes people are making to the natural world will affect evolution. To find answers, he's studying fish that live in Bahamian tidal creeks that have been fragmented by roads.

"We are looking at this on a number of different islands, and we are seeing that the fish are evolving differences really rapidly in response to human-induced changes," Langerhans says. "How predictable it's going to be across all these different species and all these different islands, I don't know yet."

Understanding that could give us insights into where the changes we are making in the natural world now could lead to many centuries from now, Langerhans adds.

"I think the point of all this is trying to figure out how the world works. Why are there so many species on this planet, and why on Earth do they look the way they look? How do we explain all this diversity?" he asks.

"And if we can get answers to those questions, it gives us information not only about the evolutionary trajectories of these species, but it also can help us understand such things as how we are affecting these ecosystems and what that's going to mean in the future."



Becky Kirkland

In his N.C. State lab, Langerhans and his students discuss the natural-selection lessons that the *Gambusia* have provided.

PERSPECTIVES
College of Agriculture and Life Sciences
Campus Box 7603
North Carolina State University
Raleigh, NC 27695-7603

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Courtesy Brian Langerhans

A CALS scientist researches the predictability of evolution through studies of the *Gambusia* fish, inhabitant of isolated blue holes – submerged caves in the Bahamas. (Story, page 15)