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# **QnAs with David B. Wake**

Paul Gabrielsen, Science Writer

The mass extinction at the end of the Cretaceous period opened up a wide range of ecological niches, and frogs were among the surviving animal groups to take advantage of the opportunity. Three lineages of frogs in particular-Hyloidea, Microhylidae, and Natatanura-were so successful in the time period following the extinction of the dinosaurs that they now represent the ancestors of 88% of living frog species. For nearly 60 years, David B. Wake, a zoologist at the University of California, Berkeley, and a member of the National Academy of Sciences, along with his wife, Marvalee Wake, has studied the evolution of reptiles and amphibians. In a recently published study in PNAS (1), Wake and his colleagues used DNA from the nuclei of frog cells, rather than the maternally heritable mitochondrial DNA, to reconstruct the timeline of frog evolution. Wake spoke with PNAS about the study.

**PNAS:** Why is it important to understand the evolution of frogs?

Wake: Frogs are a very old group of vertebrates. They've been around for 200 million years. They're a central player in most ecosystems around the world. So we're intrinsically interested in frogs as we are with other vertebrates, but with a special focus on a very highly specialized group of organisms that's been very successful through time.

**PNAS:** Why did researchers previously think that frog diversification occurred in the Mesozoic?

Wake: We do have a number of fossil frogs from the Mesozoic, and we do know that a number of living lineages go back that far. But we didn't realize that what we were looking at were a group of ancient survivors, and we missed the forest for the trees, so to speak. These three groups that we've now identified contain 88% of all living frogs.

**PNAS:** What novel genetic techniques did you use to study frog evolution?

Wake: We've known for a long time that mitochondrial DNA sequences are not the best source of information for old groups, yet we've used it because it was so much easier than going after nuclear genes. But now it's become increasingly possible to use nuclear genes. We want to know which specific genes we're using, rather than just using a huge array of unidentified nuclear DNA. We have 95 nuclear genes. Most earlier groups [had] used three or four nuclear genes, or maybe up to five or six, but by using 95 we've vastly increased our analytical power. We analyzed extant frog species. We use some sophisticated computer programs that are calibrated by fossils in what's called a relaxed molecular clock method. That's what we used to estimate the timing.

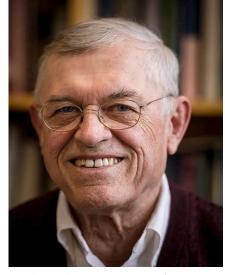
**PNAS:** What picture can you describe of frog populations both before and after the end-Cretaceous extinction?

#### Wake: There were a lot of

frogs around before the extinction event. And many of those lineages made it through. But what's the big surprise is that three of these groups are what we call "deeply nested." That is, they've gone through many rounds of splitting. They've had many opportunities to develop adaptations and specializations of all sorts and it is these deeply nested lineages that experienced this great radiative burst of evolution right at the Cretaceous–Paleogene boundary.

The loss of the dinosaurs is the hallmark event of that extinction event. It wasn't the dinosaurs themselves but everything else that disappeared that created the new ecological opportunities. Those frogs that made it through suddenly faced an array of possibilities. There are "good evolvers" and "bad evolvers" and there's a spectrum in between. If you look at, for example, the group known as leiopelmatids, they go back to the early Jurassic. There are four species from the genus *Leiopelma* that occur in New Zealand, and there are two species in the genus *Ascaphus* that occur in the Pacific Northwest. They're just fragments. They were not good evolvers, whereas these three other groups had a lot of evolutionary potential. I can't tell you exactly why. I really wish we knew why that was.

The flowering plants, the angiosperms, which include the leaf-bearing flowering trees, really took off after the extinction event too. The broad-leafed trees drop their leaves during the year and provide



David B. Wake. Image courtesy of David B. Wake.

leaf litter, which provides hiding places, a rich array of soils, and detritivores and insects as food for the amphibians. Today, arboreality in frogs is a key element of their current success.

**PNAS:** What longstanding questions in frog evolution does this study resolve?

Wake: For a long time we thought that frogs had come out of India because there were so many different groups of frogs in India. But now we realize the really stable continent has been Africa.

Among the elements of success in the frogs that did survive are arboreality and direct development, or the abandonment of the aquatic larval stage. Not any one of these is the critical issue. For example, all leiopelmatids have direct development. Having direct development is not a key to success. But having direct development is a feature that gives them access to many kinds of habitats they would not be able to occupy otherwise. Some occur largely underground in the driest and most inhospitable deserts in Australia. And they make it because they don't need standing water for reproduction. We think of amphibians as needing water during some phase of their life history, but many different lineages have become direct developers. That's part of the success of the radiation.

**PNAS:** Which of the three major lineages does your namesake *Wakea madinika* belong to?

Wake: Wakea is named for David and Marvalee Wake. It's a Madagascaran frog in the Natatanura lineage, particularly a member of the family Mantellidae, which is endemic to Madagascar and Mayotte in the Comoro islands.

**PNAS:** How did your team gather the necessary frog tissue samples?

Wake: They come from us going out [into] the field to collect them and putting them in museum collections. The Museum of Vertebrate Zoology, which I directed for 27 years here at Berkeley, has a very large frozen tissue collection now, which feeds the whole community of biodiversity scientists. That's how we built this up, by tapping into programs like the frozen tissue collections at the University of Texas, Berkeley, Harvard, the University of Kansas, and the California Academy of Sciences in San Francisco.

We're quite a diverse group. We started with the National Science Foundation's Tree of Life program in 2005. We've managed to stay together through this long period, and it was important to do this as a team because we had to assemble the very diverse group of frogs that is required.

**PNAS:** How does this work change the way we think of frogs?

Wake: I think of them as evolving right alongside the placental mammals and the great radiation of birds. We used to think of them as very ancient. I think of them now as very modern.

1 Feng Y-J, et al. (2017) Phylogenomics reveals rapid, simultaneous diversification of three major clades of Gondwanan frogs at the Cretaceous–Paleogene boundary. Proc Natl Acad Sci USA 114:E5864–E5870.

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