

Evolution, illustrated: Study captures one of the clearest pictures yet of evolution in vertebrates

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What do you get when you put together several tons of steel plates,

hundreds of mice, a few evolutionary and molecular biologists and a tiny Nebraska town near the South Dakota border?

Would you believe one of the most complete pictures ever of [vertebrate evolution](#)?

Led by Professor of Organismic and Evolutionary Biology and Molecular and Cellular Biology Hopi Hoekstra, a team of international researchers conducted a years-long study in which hundreds of [mice](#) were released into massive, custom-built outdoor enclosures to track how light- and dark-colored mice survived in light- and dark-colored habitats.

The results not only confirmed the intuition that light-colored mice survive better in light-colored habitats, and vice versa for dark-colored mice, but also allowed researchers to pinpoint a mutation related to survival, specifically that affects pigmentation, and understand exactly how the mutation produced a novel coat color. The study is described in a February 1 paper published in *Science*.

"This project has been many years in the making, and part of the inspiration for it came from the experimental evolution studies people have been doing for many years now using microbes in the lab," Hoekstra said. "The idea has been that you start with a particular population, genotype it, and then give it environmental challenges and watch how the population evolves over generations. Then you genotype it at the end and you can see, at the genetic level, what changes.

"We were interested in replicating that approach but doing it in vertebrates, and doing it in a natural environment," she continued. "And letting them evolve in habitats that—importantly—are open to predators, or at least visually hunting, avian predators."

To do it, then postdoctoral fellow Rowan Barrett (now a faculty at

McGill University) and colleagues traveled to the tiny town of Valentine, Nebraska in order to take advantage of an important natural habitat—the Sand Hills.

As early as the 1930s, Hoekstra said, it had been observed that mice living in the sand hills—a large area of contiguous sand dunes with sandy, light-colored soil—are lighter in color than those living in the surrounding areas with dark, loamy soil.

To understand what underlies those differences, Hoekstra, Barrett and colleagues came up with an ambitious plan to build a series of eight enclosures—each 2,500-meters-square, or just over a half-acre—four on the light-colored sand hills and four on the darker soil.

They then "seeded" each enclosure with 100 mice—half trapped from the sand hills and half trapped from the surrounding dark soil—after marking each with a tiny, embedded RFID tag and taking the very tip of their tails for genetic sequencing. Three months later, researchers returned and set to identify which of the mice survived.

"The idea was to start with the maximum amount of phenotypic variation, because that would give us the most power," Hoekstra said. "For this study, we asked, of those mice we started with...which ones survived after three months of natural predation?"

"So the first thing we did was to look at phenotypes, and we asked if the average color of a mouse in an enclosure changed over time" she continued. "And what was very clear was that, in the light enclosures, the average mouse became lighter and in the dark enclosures they got darker, so already we could see there had been phenotypic change."

That phenotype information, however, was only part of the story—Hoekstra and colleagues also wanted to understand whether there

may be genetic differences in the surviving populations.

"We knew that coat color was heritable, and based on our previous work, we focused on one pigmentation gene that we knew contributed to the color variation in these Nebraska populations," she said. "We sequenced the entire 200,000 base pairs of that gene...and we found a handful of [mutations](#) that were nicely correlated with survival."

The team chose to focus on one—a protein coding change that resulted in the deletion of a single amino acid from the resultant protein—which was associated with a shift toward a lighter coat color.

"That told us that mutation was associated with survival, but we wanted to know exactly what the mutation did," Hoekstra said. "We worked in collaboration with Jonathan Duke-Cohan at Harvard Medical School to do a series of biochemical analyses to show that this mutation affected the binding property of this protein in a way that leads to lighter coloration."

Further tests using gene-editing techniques, Hoekstra said, confirmed that the mutation on its own resulted in visibly lighter-colored mice.

"That was such a satisfying result—the fact that you can just look at a mouse see a difference," Hoekstra said. "You can imagine a scenario where a mutation caused a change in color, but you can only see it if you measure it with a very precise instrument, but here it was wonderfully obvious."

Going forward, Hoekstra said, there are two main paths for researchers to pursue—further investigating whether there may be other genetic differences, in pigment genes or other genes, that help mice exist either on or off the sand hills, and the other investigating change over multiple generations.

"Yes, there is a big difference between the soil color on and off the sand hills," she said. "But they also differ in vegetation and other ecological aspects, so there are likely other differences besides color at the phenotype level that we'd like to tap into as well. In that case, looking genome wide and asking if there are other regions of the genome that affect fitness will be a next step as well as looking across generations."

Ultimately, though, Hoekstra said the study offers an important—and importantly complete—picture of how evolution works in mammals.

"What I like about this study is it starts at the organismal level and goes all the way down to a mutation, and understanding how, precisely, that mutation leads to a change in phenotype and how that change leads to survival differences in the wild," she said. "So I think this is a very satisfying illustration of the full process of evolution, from the ecological consequences of these phenotype changes down the molecular details."

More information: "Linking a mutation to survival in wild mice" *Science* (2019). [science.sciencemag.org/cgi/doi ... 1126/science.aav3824](https://science.sciencemag.org/cgi/doi/10.1126/science.aav3824)

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