

## First Steps: Scientists launch evolutionary study to explore the origins of fish that walk

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A new research collaboration with support from the National Science Foundation, or NSF, begins an unprecedented study of walking cavefish to better understand the "fin-to-limb" transition that enabled the first vertebrates to walk on land more than 350 million years ago. This new research collaboration between LSU, New Jersey Institute of Technology, or NJIT, and University of Florida is set to launch the first evolutionary study of the unique pelvic structure and walking mechanics of the waterfall-climbing blind cavefish, or Cryptotora thamicola—the only living species of fish known capable of walking on land with a similar motion as four-limbed vertebrates, or tetrapods, which include mammals, reptiles and amphibians.

Researchers say the project's unprecedented, comprehensive study of the cavefish's vertebrate-like pelvic girdle and phylogeny may provide a new window into the evolution of appendages, pelvis and vertebral column that ancient tetrapod fishes needed to transition to terrestrial life during the Devonian period, between 350 and 400 million years ago.

"Combining robotics, genomics and CT morphological examinations, this collaboration could help us visualize evolution in a brand new light," said Co-principal investigator Prosanta Chakrabarty, who is the LSU Museum of Natural Science Curator of Fishes and an associate professor in the LSU Department of Biological Sciences. "With support from the National Science Foundation's Understanding the Rules of Life initiative, we are proud to embark on this landmark study to identify the genomic, morphological and mechanical qualities that enable <u>fish</u> to



walk on land."

The research team at LSU, led by Chakrabarty, will conduct genomic sequencing of the cavefish and its relatives to determine convergent genetic markers known to be involved in pelvic girdle formation in other walking vertebrates.

"Evolutionarily, there are a number of different ways that other species may have approached the water to land transition, but the physics and the fundamental properties of land-walking that the cavefish has had to overcome by developing their pelvic structure, those things have not changed in 350 million years," said Brooke Flammang, the project's lead principal investigator and assistant professor of biological sciences at NJIT. "This study, for the first time, will provide the opportunity to study and measure a fundamental mechanism that was key to this major evolutionary transition."

NSF announced the project, titled "A Phylogenomically-Based Bioinspired Robotic Model Approach to Addressing the Evolution of Terrestrial Locomotion," will receive a four-year, \$997,510 grant to examine the phylogenomic relationships, morphology, biomechanics and walking performance of the <u>blind cavefish</u> and its relatives, from an evolutionary perspective.

"Our goal is to help answer how and why we often see repeated evolution of similar anatomies in different parts of the Tree of Life," Chakrabarty said. "To do so, we apply molecular techniques at my lab at LSU to better understand the potential genetic convergence and evolution of the phenotypic anatomy being studied at the University of Florida and the functional anatomy being examined at NJIT."

Until recently, all living fishes were thought to lack the tetrapodal characteristics necessary for studying the physics of quadrupedal



walking by fishes in a terrestrial environment. In 2016, Flammang and fellow NJIT faculty-researcher Daphne Soares initially observed that the two-inch-long cavefish shared morphological features associated with terrestrial vertebrates, identifying that the fish utilized a robust pelvic girdle attached to its vertebral column in order to walk and climb waterfalls with a salamander-like gait in its native habitat, the Tham Maelana and Tham Susa karst cave systems in northern Thailand.

"There have not been many models or preserved fossil records available to help us understand how the pelvis evolved and formed in the first tetrapods that developed appendages from fins to walk on land," Flammang said. "These cavefish may not necessarily offer a living analog of what ancient tetrapod fish used to look like, but in better understanding the evolution of the pelvis in this cavefish, we may have a window backwards into the physics of early tetrapodal weight-bearing limb support necessary to walk on land."

The research collaboration will aim to identify the morphological, genomic and mechanical qualities of walking cavefish that may have enabled the first fishes to walk on land. University of Florida researchers, led by Larry Page, Florida Museum of Natural History Curator of Fishes and affiliate professor of biology, will lead the study's morphological study and collection effort from as many as 238 fish species that share the same family as Cryptotora thamicola, known as the Balitoridae, for comparative analysis. The morphological analysis will reveal how as a generality, functional phenotypic adaptations can be formed by genomic changes.

Flammang's lab will use phylogenomic, morphological and experimental biomechanical data to create a biorobotic model of the fish. The robotic model will be used to examine possible instances of evolutionary convergence and answer questions on the effect of pelvic morphology on walking performance in extinct fin-to-limb transitional species forms, as



well as modern forms.

"This will be the first time we have a phylogeny of this family of fishes that includes this unusual <u>cavefish</u>, and it also may be the first case of anyone using biorobotics to model a major evolutionary transition," Flammang said. "Once we have a working model of the fish and the evolution of the pelvis based on what we know of these fishes, we can apply the model to other organisms that we have fossil evidence for their morphology to get an idea of how they may have walked at stages in their evolutionary history."

## Provided by Louisiana State University

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