Chemical Signals of Molecular Preservation Q&A with Dr. Paul Ullmann

On Sept. 23, Ullmann et al published **"Molecular tests support the viability of rare earth elements as proxies for fossil biomolecule preservation,"** in the journal *Scientific Reports*.

A paleontologist, Ullmann, an assistant professor in the Department of Geology within Rowan's School of Earth & Environment, wrote that the key "take home" message of the study was that researchers used three molecular biology techniques to document preservation of the original protein collagen in a fossilized bone that experienced minimal chemical alteration during fossilization. Their work establishes a new link between the traditionally separate fields of geochemistry and molecular paleontology, and demonstrates for the first time that elemental chemistry can be successfully used to identify the likelihood of a fossil bone to still contain original molecules.

The team collected the fossils from a vast South Dakota dinosaur bonebed over three weeks in the summer of 2012.

Read more about how the research team formulated its objective and devised a way to use rare earth elements as indicators of molecular preservation potential in fossilized bone.

Q: Who took part in the excavation?

A: Ullmann led a collaborative team of six scientists, graduate students, and an undergraduate student from three institutions: Drexel University in Philadelphia, Concordia College in Moorhead, Minn., and the Standing Rock Sioux Tribe Paleontology Department in Fort Yates, North Dakota.

Q: What was the research question?

A: Can the rare earth element composition of a fossil bone be used to identify its potential to contain original molecules?

Q: How did the authors test this? What did the authors do?

A: Ullmann's team previously documented the chemical composition of fossil bones of a duck-billed dinosaur called *Edmontosaurus* from South Dakota, and based on those results they predicted that the bones should be favorable candidates for molecular preservation.
In the present study, they directly tested that prediction: Ullmann and his team performed protein extractions and three molecular biology tests on one of the fossils. They discovered that the bone does indeed preserve original protein, as predicted, validating the use of rare earth elements as a screening tool prior to molecular testing.

Q: What are rare earth elements (REEs)?

A: The rare earth elements are chemical elements 57 through 71 of the periodic table of elements, and they are often depicted in a separate row along the bottom of the periodic table. They include the metals lanthanum through lutetium, and are called "rare earth" because they occur in very small amounts in most rocks and waters on Earth. They also have economic uses in computer microchips, rechargeable batteries, magnets and fluorescent light bulbs.

Q: How are rare earth elements connected with bone fossilization?

A: After an animal dies, cells and soft tissues in bones, including blood vessels and structural protein fibers, begin to decay and rot away to leave behind empty spaces. Once the bone is buried in a natural environment, groundwater flows through those spaces and brings with it rare earth element ions from the surrounding rocks. Bone mineral is very reactive, so it absorbs these rare earth elements from groundwater as it passes through, and that process contributes to the bone's alteration to a stable fossil. As a result, the rare earth element composition of a fossil bone includes signatures of the chemistry of the environment in which it was buried and provides a direct record of how intensively a bone has been chemically altered through fossilization.

Q: Why did you expect rare earth elements to be connected with the preservation of molecules in fossils?

A: Since rare earth elements record information about chemical alterations that took place during fossilization, they can be used to predict if fossilization conditions were favorable to the preservation of biomolecules. For example, if one fossil is found to be less chemically altered than another, then the less altered fossil is likely to have experienced less harsh chemical conditions during fossilization, which in turn makes it more likely to retain some of its original molecules.

Q: What fossils were analyzed, and where were they collected?

A: Over 300 bones of the Cretaceous duck-billed dinosaur Edmontosaurus annectens were collected from a bonebed called the Standing Rock Hadrosaur Site, or SRHS for short, on land owned by the Standing Rock Sioux Tribe in Corson County, S.D. Of these 300+, Ullmann selected nine limb bones for rare earth element analyses, the results of which were presented in a previous study. In the present study in *Scientific Reports*, one of those nine bones was further tested for preservation of the protein collagen I.

Q: What is molecular paleontology?

A: Molecular paleontological research includes the search for, identification of, and interpretation of original biologic molecules in fossils. This includes ancient DNA, proteins, carbohydrates and lipid (fat) molecules. This is a growing field of science, and the molecular study of dinosaurs was in large part initiated by co-author Schweitzer's papers in Science in 2005 and 2009 which documented for the first time the preservation of original proteins in extinct dinosaurs *(Tyrannosaurus rex* and *Brachylophosaurus canadensis*, respectively).

Q: What molecule or molecules were recovered from the fossil bones?

A: The authors conclusively identified the bone structural protein collagen I.

Q: Was ancient DNA found?

A: No. Ullmann and his team identified preservation of original protein in the fossil bone, not DNA. They did not look for DNA.

Q: How are you certain you found dinosaur protein?

A: Ullmann and co-author Voegele conducted protein extractions followed by three molecular tests for the presence of biologic molecules in the *Edmontosaurus* bone studied here. First, gel electrophoresis tests were performed to determine whether any organic molecules were present in the fossil bone, and to gain an idea of their relative size. These results were positive, suggesting organics were present in the bone. Next, two immunological tests were performed to test specifically for the protein collagen I: immunofluorescence and enzyme-linked immunosorbant assays (ELISAs). These tests use antibodies created by an animal's immune system to recognize the presence of specific target molecules, in this case collagen I. ELISAs successfully identified the presence of the protein collagen I, and immunofluorescence tests constrained this positive result to come from fibrous tissue masses isolated from the bone via treatment with a mild acid. Sediment and solution controls that were run at the same time as the protein tests all came back negative, demonstrating the collagen pertains to the fossil and does not originate from any type of external contaminant.

Q: Why are discoveries of ancient biologic molecules like proteins important?

A: Discoveries of ancient biologic molecules have many diverse uses in paleontology and other scientific disciplines. First, ancient molecular sequences can be used to independently test hypotheses of evolution, such as how extinct animals like dinosaurs are related to living animals. Second, discoveries of the same molecule from multiple fossils also can provide insights into how that molecule is evolving over time, which can help predict how biologic molecules may evolve in the future as animals respond to today's ongoing global climate change. Third, since only small fragments of ancient molecules survive the fossilization process, scientists have had to adapt instrumentation and protocols for use on fossil specimens, which continues to advance the resolution of these analyses and decreases the amount of sample required to perform physical and chemical analyses of biologic samples. Those analytical advancements are likely to see applications in the future in everything from diagnostic medicine to materials engineering (e.g., biofabrics).

Q: Have any other analyses been performed on the bones?

A: Yes. Ullmann previously published three papers on these fossils: a 2017 paper in the journal *PALAIOS* discussed how the dinosaurs died and were buried; an early 2019 paper in Cretaceous Research documented the preservation of original bone cells, blood vessels and protein matrix within these bones; and a late 2019 paper in the geochemistry journal *Geochimica et Cosmochimica Acta* presented detailed studies of the chemical history of these bones as they became fossils.

Q: Is the team actively exploring molecular preservation in other fossils?

A: Yes. Ullmann is conducting multiple projects exploring the fossilization history of dinosaur and other reptile bones from across the United States in an effort to characterize environmental and geochemical conditions conducive to molecular preservation in fossils.

Q: What is the utility of the author's findings to other paleontologists?

A: Ullmann and colleagues show that rare earth element chemistry of a fossil bone can be successfully used to identify whether or not it is likely to contain original (ancient) molecules. This gives paleontologists a criterion for narrowing down a set of fossils to those which should be the most productive targets for analysis, which will ultimately lead to additional molecular discoveries from fossil bones. Ultimately, new molecular discoveries made by applying this screening tool will yield novel insights into the biology, physiology, and evolution of the extinct animals they derive from.

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Rowan University is a Carnegie classified national doctoral research institution dedicated to excellence in undergraduate education. Founded in 1923, Rowan University has evolved from a teacher preparation school to a comprehensive research university that's ranked by U.S. News & World the best national public research universities in the nation. The historic $100 million Rowan gift in 1992 put the institution on the fast track, fostered the creation of the College of Engineering and enabled ambitious growth. Nearly 96 percent of Rowan students are New Jersey residents. In all, the student body hails from 41 states and 35 countries.

About research

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Rowan University's School of Earth & Environment launched in 2015 to provide students pathways into some of the most exciting and important careers in the modern workforce, to gather outstanding faculty who are globally recognized leaders and to welcome non-academics into our community. Understanding the interactions between Earth systems, life on our planet and human society is essential to a flourishing future for all of humanity.

The School of Earth & Environment at Rowan University includes the departments of Environmental Science; Geography, Planning & Sustainability; and Geology.

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