

Signatures from the Past

**How
isotope
research digs
deep to solve
mysteries from
ancient Earth
and beyond.**

By Lori Dajose (BS '15) 🌟 Illustrations by Charis Tsevis



John Eiler used isotope studies to measure the body temperature of dinosaurs.

It was Thanksgiving Day 2015, and geochemist John Eiler was sitting alone in an airport in Bremen, Germany, eating goose—the closest thing he could find to the turkey he would have been having back home with his family in Sierra Madre. He had ventured to Bremen, an ancient port city of industrial infrastructure, on a quest to realize his dream of creating a powerful machine that could see tiny variations in the abundances of rare isotopes and map their positions within a single molecule.

All matter is made up of chemical elements. The individual atoms of a particular element can vary in the number of neutrons in their nuclei; these variants are called isotopes. For decades, existing instruments called mass spectrometers had been effective at measuring the ratios of different isotopes—each of which have a different atomic mass but the same elemental properties—of atoms and of the atoms within a few simple molecules. These ratio measurements had enabled major discoveries about the solar system and Earth’s ancient history, but they had also missed the vastly more comprehensive and potentially useful information encoded in the isotopic structures of more complex molecules. Eiler knew that with a more powerful and specialized machine that could interrogate these isotope structures, isotopes could be used to answer an even broader set of questions about Earth, the wider universe, and the human body.

Isotopes act as chemical fingerprints, enabling a forensic-like analysis of materials that can answer an array of surprisingly diverse questions, like what extinct giant sloths liked to eat and where asteroids formed in the cold expanse of outer space. The technology Eiler spent years designing and developing would go on to break new ground, allowing researchers to unlock even more valuable data. “I felt like it was going to happen, but I don’t think anybody else thought that it would happen,” he says of the arduous process.

Eiler’s work builds upon a long history of Caltech researchers who have pushed the frontier of isotope geochemistry since the 1950s, using isotopic signatures to discover Earth’s age, analyze lunar samples obtained by the Apollo missions, and understand ancient climate records preserved in natural environments. Now, a new generation of Institute faculty, including geobiologist-paleontologist Julia Tejada and geochemist François Tissot, is adding to that legacy by pioneering new technical advances and practical applications that have led to a wide variety of breakthroughs, including better ways to detect osteoporosis in humans, a more detailed understanding of the evolution of life on Earth, and how the chemical building blocks of all living things got here.

A Paleontologist’s New Tool Kit

On her annual expeditions to the hot, humid Amazon rainforest, Julia Tejada wields the traditional tools of a paleontologist—pickaxes, brushes, hand lenses, the ability to ignore swarms of mosquitos—with ease while unearthing ancient animal fossils. But she is not a traditional paleontologist. Tejada’s tools back in the lab enable her team to peer at the atomic-level isotopic variations in fragments of fossil samples. “You don’t need isotopes to look at a saber-toothed tiger skull and know that it ate meat; just look at the morphology of its

Did You Know?

Julia Tejada is only the second paleontologist to join the Caltech faculty. Learn more about the first, Chester Stock, and a feline fossil from his collection still on display at the Institute, on page 13.

Julia Tejada uses isotope geochemistry to study the fossils of ancient animals and learn more about their lives.



teeth,” she says. “But to answer deeper, subtler questions about how ancient life evolved on Earth, you need to look at the molecular level.”

Isotope geochemistry allows Tejada to study ancient environments like lush forests and oceans even though they are long gone. This is because their chemical record can still be found in the fossils of creatures that roamed Earth millions of years ago, such as bear-sized sloths and giant penguins. These early environments contained water, air, and nutrients with isotopic ratios that differ from those of today and which left distinctive signatures in ancient fossils. Tejada studies isotopes in these fossils to uncover stories about the past; in particular, she examines extinct sloths (see cover) that could grow to be almost 10 feet tall and were a major component of ancient ecosystems.

“More than 90 percent of organisms that have ever lived on our planet are now extinct,” says Tejada, who



joined the Caltech faculty in 2023. “To understand how the evolution of life led us to where we are today, we have to look deep back in time. In the absence of fossil plants, biomarkers preserved in soil or incorporated into bones, teeth, or hair can tell you something about the environment in which an organism lived millions of years ago.”

Most elements on the periodic table exist in multiple isotopic variations. For example, the majority of carbon atoms have six neutrons and six protons in their nuclei, earning the name carbon-12 or C-12. But slightly more than 1 percent of all carbon atoms have seven neutrons (and six protons) and are thus dubbed carbon-13. Extra neutrons make atoms heavier, which causes them to behave slightly differently in chemical reactions. These variations in weight and behavior can be detected in the lab. Using mass spectrometers, researchers can measure isotopic ratios to draw conclusions about an object’s history.

While Tejada’s laboratory contains the modern tools to make measurements at the atomic level, it also seems to double as an animal-skeleton emporium. During a hot morning this past July, she pulled large bones out of cardboard boxes to show students, deftly explaining the differences between a pelvis and a femur. These particular bones, she noted, were not fossils; they came from a modern cow. Tejada will use them to teach the students how to properly cut and treat bone samples before they move on to handling more precious fossil samples.

Tejada’s lab can analyze these older specimens and make startling discoveries with a mere whisper of material. Her work has revealed surprising insights that may not be evident from morphology alone. For example, a particular ancient sloth was presumed to be a herbivore due to the shape of its preserved teeth. By studying the isotopes present in specific amino acids within the sloth fossil, Tejada proved that the animal also ate meat, overturning the original hypothesis.

In this way, isotope geochemistry helps underscore the old maxim, “You are what you eat.” For example, elements you consume become embedded within your bones, teeth, and hair. But the power of isotopes is not limited to analyzing nutrition in living beings; many chemical processes impact isotopic ratios. Certain isotopes of lead are only produced as part of the decay of uranium atoms over millions of years. The more of this “radiogenic” lead in an object, the longer the object has been around. The late Caltech geochemist Clair Patterson used this logic, along with measurements of the lead isotope ratios in ancient rocks, to measure the age of our planet: 4.6 billion years, certainly enough time to evolve myriad sloths.

Digging Deeper

Rainwater in the Rocky Mountains is isotopically lighter than in California because as clouds travel across land from the ocean, heavier isotopes are rained out, leaving a higher abundance of light water by the time the clouds reach the interior land. In this way, the water you drink carries a signature of where it came from.

A History of Discovery

Before the 1950s, isotope research in the United States focused primarily on isotopes of a certain subset of radioactive elements, like plutonium and uranium, that could power nuclear weapons. After World War II, scientists who honed their skills on the Manhattan Project found themselves looking for less destructive ways to apply their knowledge. In the '50s, several young scientists who had studied how to translate isotope science into the study of ancient Earth came to Caltech.

These geochemists turned Caltech into a powerhouse in the geological and planetary sciences. After Patterson created one of the nation's first clean room laboratories in the process of measuring Earth's age, he used his expertise to campaign for the removal of lead from gasoline, reducing air pollution. Samuel Epstein, who was invited to Caltech by Harrison Brown, founder of the Institute's geochemistry program, used isotopes to study the Antarctic and Greenland ice sheets and learn about the origin of meteorites, tektites, and lunar rocks and minerals. In a laboratory fondly nicknamed the "Lunatic Asylum," lunar expert Gerald Wasserburg built the first-ever digital mass spectrometer and used isotopes from lunar samples to study cataclysmic collisions between asteroids and the Moon. Due to these scientists and other luminaries in the field, the Institute's technology and laboratories became top of the line, with researchers accessing the latest mass spectrometers for their discoveries.

When Eiler arrived at Caltech in 1994, he found himself in awe of these famed geochemists and others. "They had really big personalities, and they trained virtually everybody who was influential in the field at that period," he says. "My first impression was, 'I'm not ready for this. These people are on a completely different level!'"

Still, Eiler soaked up their insights like a sponge. After a few years of attending daily seminars and developing what he calls "good taste in problems," he proposed a new direction for isotope research that would ultimately define his career. It was driven by a simple question: What if, instead of simply measuring how much of a given isotope was in a sample, you could also measure where an individual isotope was located in the structure of each molecule?

Eiler knew that if you could understand isotopic positions within molecular structures, the questions you could answer would be extremely wide ranging. It amounts to the difference between having aggregate data about a large group of people versus knowledge of each individual's specific qualities. Suddenly, otherwise identical molecules become unique, like a fingerprint.

Consider an everyday molecule of sucrose, the main component of white sugar: One molecule of sucrose contains 12 carbon atoms, 22 hydrogen atoms, and 11 oxygen atoms arranged in two connected hexagonal loops with protruding spiky branches. Each of these atoms could be a different isotope—perhaps a heavy oxygen atom sits on the outside edge of the molecule, or perhaps close to its center. Maybe there are two, or more, heavy isotopes of hydrogen, carbon, or oxygen—or all three. When all these possible combinations are considered, sucrose must exist in quadrillions of isotopic variations, each pointing to the unique conditions under which they formed.

"The isotopic structures of molecules—that is, the numbers and spatial organizations of the rare isotopes they contain—can be measured on literally any compound known to science, and they record a vast amount of detailed information about how and where that molecule formed and what it has experienced since its formation," Eiler explains. "The ability to probe these data brings the complexity of something like genomics to the study of even simple, mundane-seeming molecules. This is something that can be used to understand the formation of molecules in interstellar space, the evolution of life on Earth, the detection of life on Mars or other planets, or metabolism and disease in the human body."

Eiler knew he would need a machine more sensitive than existing mass spectrometers to accurately examine a large variety of molecules and differentiate their isotopic signatures. Unfortunately, such a tool did not exist—yet.

Star Stuff

While Tejada and Eiler use isotopic geochemistry to examine Earth's past environments, their next-door colleague in Caltech's North Mudd Laboratory, geochemist François Tissot, started his career by using isotopes to peer even further into the past—to the beginning of the solar system itself.

Tissot keeps pieces of ancient meteorites safely locked in his office and will readily pull them out to show to any visitors who may be interested. Shiny white geometric chunks within the rocks are compounds of calcium and aluminum, the oldest remaining objects in the 4.6-billion-year-old solar system.

Looking at these rocks, it is hard to imagine that the elements in our own bodies also have the same far-off origins—but it is true. The calcium atoms in our bones, along with all other elements on Earth, were originally forged in the heat of stars—indicating we are not only what we eat, but we are also "made of star stuff," in the words of the late astronomer Carl Sagan. While we share

common origins with all matter on Earth, isotopes are able to tell tales of the journeys that atoms and molecules have taken to reach their present-day forms.

Take Hadean zircons, for example—tiny crystals from the early formation of Earth, preserved over 4 billion years, and the only remaining solids from the earliest crust of the planet. Tissot wanted to use zircons to understand Earth's primordial environment. In the absence of any rock from this time period, finding hard evidence of what Earth looked like is a true challenge, and many varying theories about its earliest days have been proposed. For instance, it had previously been hypothesized that our planet was so hot and radioactive at this time that nuclear fission spontaneously occurred on the surface. The isotopic composition of uranium in zircons could confirm or refute this, but the samples of zircon available were far too minuscule to use traditional techniques. So, in 2018, Tissot invented a way to probe the isotopic ratio of uranium inside a single crystal of zircon, and he used that to show there is no evidence supporting the hellish vision of an early Earth covered in natural nuclear fission reactors.

Tissot had always wanted to find a way to apply the knowledge and expertise he gained from studying

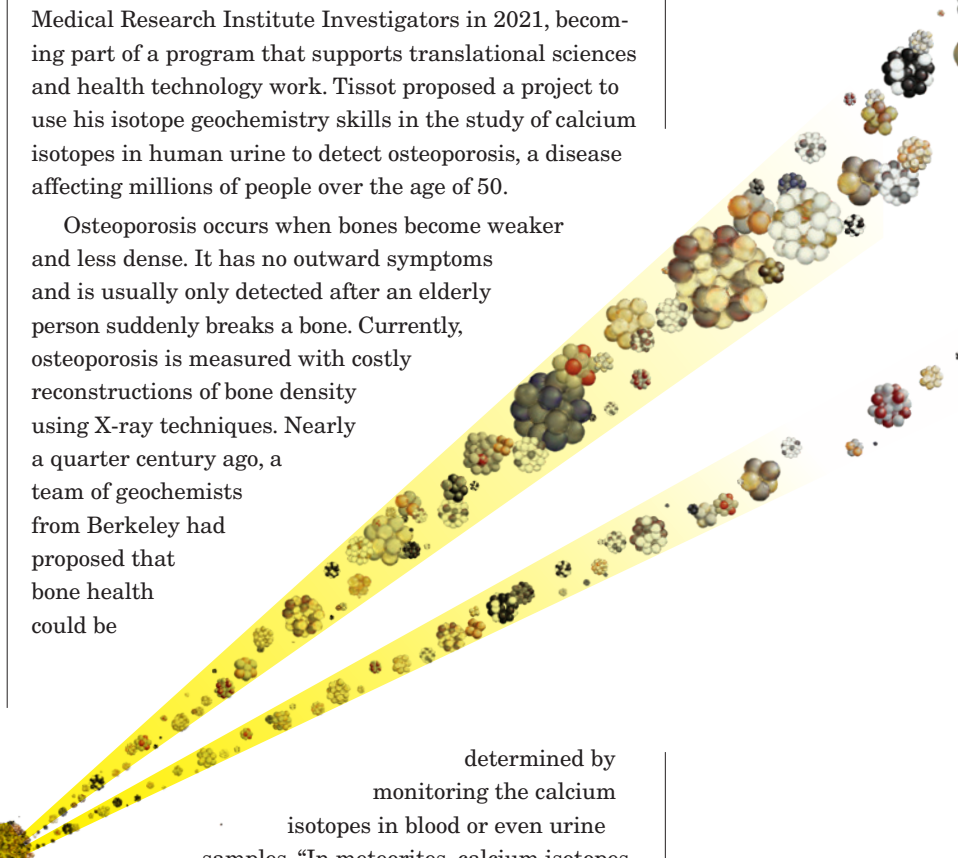
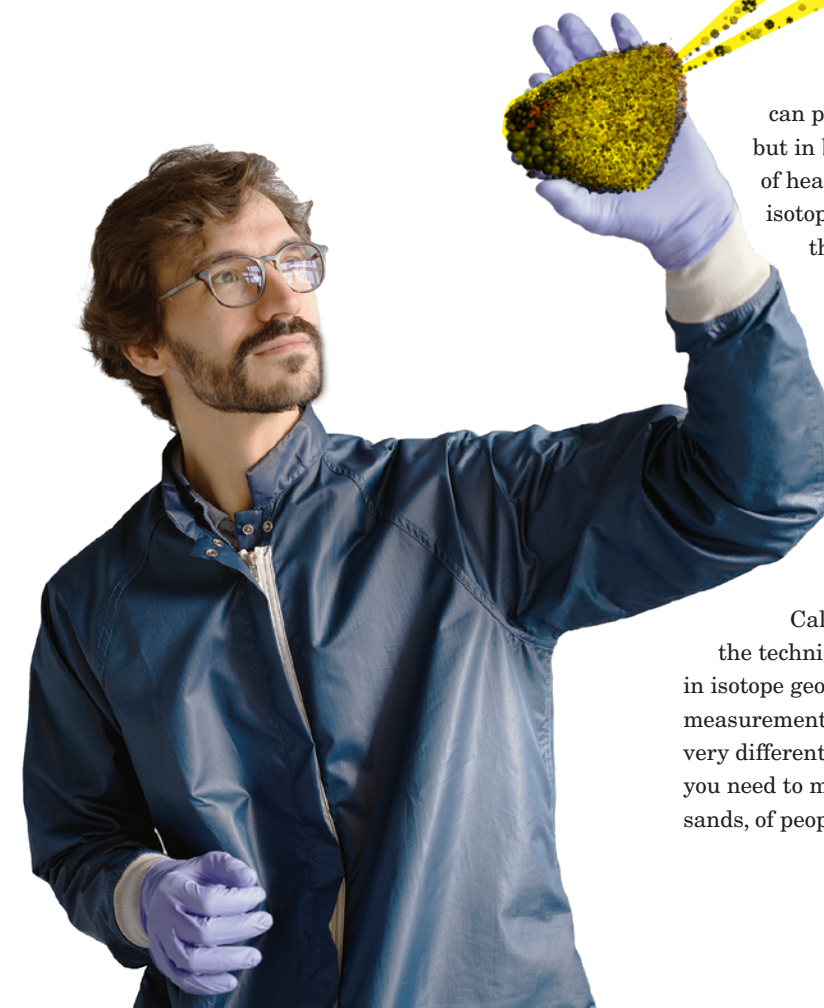
meteorites and terrestrial samples to help humankind, and he got that opportunity after joining the Institute faculty in 2018. He was named one of Caltech's Heritage Medical Research Institute Investigators in 2021, becoming part of a program that supports translational sciences and health technology work. Tissot proposed a project to use his isotope geochemistry skills in the study of calcium isotopes in human urine to detect osteoporosis, a disease affecting millions of people over the age of 50.

Osteoporosis occurs when bones become weaker and less dense. It has no outward symptoms and is usually only detected after an elderly person suddenly breaks a bone. Currently, osteoporosis is measured with costly reconstructions of bone density using X-ray techniques. Nearly a quarter century ago, a team of geochemists from Berkeley had proposed that bone health could be

determined by monitoring the calcium isotopes in blood or even urine samples. "In meteorites, calcium isotopes can provide clues about the early solar system, but in bones, they can serve as a useful marker of health," Tissot notes. "In particular, calcium isotopes in bone tend to be slightly less heavy than those found in blood, tissue, and muscle. In osteoporosis, this light calcium leaks out of the bone and into the bloodstream, altering the blood's isotopic ratios and ultimately becoming detectable in urine." While the rationale is simple to understand, the potential of the approach had seldom been put to the test.

To demonstrate the medical applications of this research, Tissot and his team first solicited participants from the Caltech community to provide urine samples so the technique could be refined. "The techniques used in isotope geochemistry are very good at making precise measurements of tiny purified samples," he says. "But it's very different to transfer to a medical application where you need to measure samples from hundreds, if not thousands, of people in a reasonable time frame."

François Tissot studies isotopes in meteorite fragments to learn more about the origins of our solar system.



Next, his lab scaled up the project with a nearly 300-person population survey to see whether or not calcium isotope levels vary demographically or over short periods of time in healthy people. As a result of their findings, the Tissot lab has developed procedures for handling more than 100 samples in a single week, indicating that their techniques could one day be scaled up and implemented in clinical settings that manage larger volumes. “We can now do measurements that are unparalleled in precision and at such a rate that large-scale clinical studies are possible. We’re starting to design them now,” Tissot says.

In addition to the osteoporosis project, Tissot remains connected to his planetary science roots. Recently, Tissot gave a lecture discussing how samples returned from Venus could tell us at what point in their evolution Earth and its twin diverged so dramatically. “Whether it’s in the human body or outer space, isotopes respond to the same general rules no matter the system,” he says. “If the question is interesting, the isotopes allow you to study it.”

Learn more about how sample return work continues to play an important role in Caltech’s Division of Geological and Planetary Sciences.



Eiler’s Machine

In 2003, Eiler began working with LA-based Thermo Fisher Scientific to achieve his goal of nailing down the exact locations of isotopes within atoms in order to unlock vast amounts of molecular information. The trick was convincing the manufacturer that new types of machines designed for this purpose would not only function, but that they would be used by other researchers. “I think they thought I was crazy,” Eiler recalls.

Although the first generation of this new mass spectrometer was capable of studying only a small fraction of the chemical compounds he hoped was ultimately possible, Eiler produced some preliminary findings and began to give talks around the world about the machine’s potential for breakthroughs. As knowledge of the instrument and its use grew, and with as many as 50 other labs signaling their interest in the technology, Thermo Fisher became convinced of the utility in developing the product, which would eventually become known as the Orbitrap mass spectrometer.

Eiler filed a patent and continued to work through generations of increasingly more capable designs of the Orbitrap with Thermo Fisher engineers, visiting the company’s factory in Germany multiple times. The Orbitrap’s capabilities expanded, enabling researchers to examine the isotopes in more complex molecules. And Eiler continued to demonstrate its value through his own research. In 2011, his team showed that isotopes in fossilized dinosaur eggshells could be used to directly measure, for the first

time, the body temperatures of ancient dinosaurs. In the years since, Eiler’s lab has made similar breakthroughs in the understanding of the ancient climate on Mars and the origins and fates of methane and other organic molecules in the natural environment.

In the last decade, Eiler’s dream of creating an atom-by-atom view of a wide range of molecules has been realized. Thanks to the Orbitrap, more precise measurements of isotope ratios have been made possible—even in small samples. This ability could prove crucial in planning future missions to study samples returned from other bodies in the solar system like asteroids and Mars.

Orbitrap mass spectrometers are now up and running in labs around the world, and multiple versions of the machine are now available. They are making a difference on campus as well. In addition to the work Tejada and Tissot are doing, collaborators across Caltech are using the Orbitrap in a wide variety of studies: to look at the degradation of plant litter in soil, the “prebiotic” chemistry that enabled the emergence of life, and more. Eiler’s team even uses it to help detect steroidal doping in elite athletes, funded by the research collaborative Partnership for Clean Competition. “Molecules of synthetic testosterone look identical in shape to the testosterone naturally produced by your body, but not when you really scrutinize down to the isotopic level,” Eiler explains. “We can take blood samples and see if the testosterone all looks the same, or if there are two different kinds—which would indicate that someone was doping.”

The Orbitrap’s diverse applications are the reason Eiler dedicated so much time to building the technology. “You don’t ask the first person to invent a telescope what star they’re looking for,” Eiler says. “This is to look at absolutely everything. If there’s a molecule, and you have a question about it, we can do something to answer it.”

Eiler’s successful journey to expand the realms of possibility builds on a legacy of geochemistry excellence at Caltech that helped draw both Tejada and Tissot to Pasadena. “John is the man,” Tissot says. “It’s inspiring to be surrounded by people who are legends in their fields.”

John Eiler is the Robert P. Sharp Professor of Geology and Geochemistry and the Ted and Ginger Jenkins Leadership Chair of the Division of Geological and Planetary Sciences. His work is funded by the National Science Foundation, among others.

Julia Tejada is an assistant professor of geobiology and a William H. Hurt Scholar. Her work is funded by the Shurl and Kay Curci Foundation, among others.

François Tissot is a professor of geochemistry and a Heritage Medical Research Institute Investigator. His work is funded by the Heritage Medical Research Institute, a Packard Fellowship, and the National Science Foundation.

An Intriguing Red Planet Rock

A sample discovered by the Perseverance rover offers hints that Mars may have hosted microbial life billions of years ago.

By DC Agle

A vein-filled rock nicknamed “Cheyava Falls” discovered by the Mars Perseverance rover on the northern edge of Jezero Crater in July 2024 contains fascinating traits that may indicate possible signs of ancient microbial life.

The rock, which measures 3.2 feet by 2 feet, exhibits chemical signatures and structures that could have been formed by life billions of years ago when the area being explored by the rover contained running water. The rock was collected on July 21, 2024, as the rover explored an ancient river valley called Neretva Vallis that measures a quarter-mile wide and was carved by water rushing into Jezero Crater long ago. Multiple scans of Cheyava Falls by the rover reveal it contains organic compounds. While such carbon-based molecules are considered the building blocks of life, they also can be formed by nonbiological processes.

“Cheyava Falls is the most puzzling, complex, and potentially important rock yet investigated by Perseverance,” says Ken Farley, Caltech’s W. M. Keck Foundation Professor of Geochemistry and Perseverance project scientist at JPL, which Caltech manages for NASA. JPL manages the Mars 2020 mission. “On the one hand, we have our first compelling detection of organic material, distinctive colorful spots indicative of chemical reactions that microbial life could use as an energy source, and clear evidence that water—necessary for life—once passed through the rock. On the other hand, we have been unable to determine exactly how the rock formed and to what extent nearby rocks may have heated Cheyava Falls and contributed to these features.”

Running the rock’s length are large white calcium sulfate veins, between which lie bands of material whose reddish color suggests the presence of hematite, one of the minerals that gives Mars its distinctive rusty hue. When Perseverance took a closer look at these red regions, it found dozens of irregularly shaped, millimeter-sized off-white splotches, each ringed with black material akin to leopard spots that contain both iron and phosphate.



NASA’s Perseverance Mars rover used its Mastcam-Z instrument to capture this view of the Cheyava Falls rock sample within the rover’s drill bit.

“To fully understand what happened ... we’d want to bring the Cheyava Falls sample back to Earth, so it can be studied with the powerful instruments available in laboratories.”

“These spots are a big surprise,” says David Flannery, an astrobiologist and member of the Perseverance science team from the Queensland University of Technology in Australia. “On Earth, these types of features in rocks are often associated with the fossilized record of microbes living in the subsurface.”

Spotting of this type on sedimentary terrestrial rocks can occur when chemical reactions involving hematite turn the rock from red to white. These reactions, which may serve as an energy source for microbes, can also release iron and phosphate, which cause black halos to form. In one scenario posed by the Perseverance science team, Cheyava Falls was initially deposited as mud with organic compounds mixed in that eventually cemented into rock. Later, a second episode of fluid flow penetrated fissures in the rock, enabling mineral deposits that created the large white calcium sulfate veins seen today and resulting in the spots.

“To fully understand what happened ... we’d want to bring the Cheyava Falls sample back to Earth, so it can be studied with the powerful instruments available in laboratories,” Farley says.