ALS SCIENCE HIGHLIGHT

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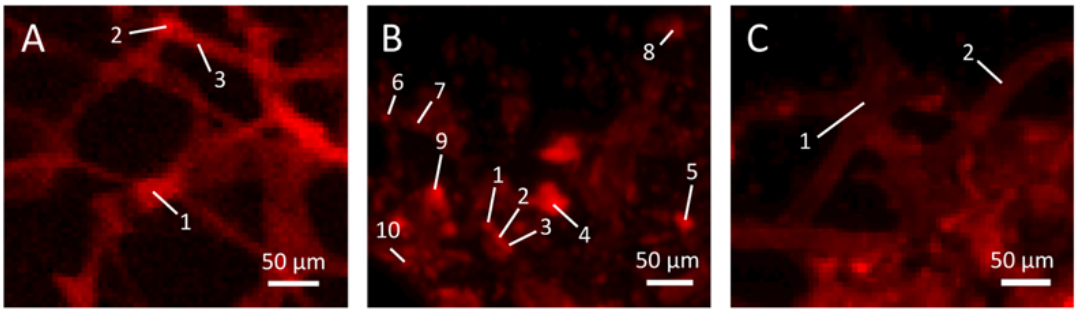
Iron is Key to Preserving Dinosaur Soft Tissue



These micro x-ray fluorescence maps of ostrich, brachylophosaurus, and tyrannosaurus rex vessel tissues (locations of analysis identified by white numerical labels) illustrate the intimate association of iron with each vessel tissue.

Researchers studying organic material from dinosaur bones have been able to show that the organic material in the samples contained original soft tissue material from Mesozoic dinosaurs. The x-ray techniques at the ALS were key to showing a possible mechanism for this unexpected preservation—iron nanoparticles associated with dinosaur blood vessels were identified 09at the ALS. Researchers hypothesized that the iron had come from dinosaurs' blood and muscle cells during decay, and were able to identify iron-facilitated reactions that contribute to preservation. If these reactions occur in other organisms that end in the fossil record, similar preservation may allow the identification of molecular evolutionary relationships, rates and direction of evolutionary change, and eventually the characterization of other traits that have, until now, remained beyond scientists' grasp.

Back in 2005, researchers observed soft, flexible, transparent vessels, fibrous matrix, osteocytes, and intravascular material in the demineralized bones of a *Tyrannosaurus rex.* These soft tissue components bound antibodies specific for proteins commonly found associated with the same components in surviving remains, and yielded amino acid sequences consistent with proteins from similar materials recovered from existing bones. This was unexpected, because conventional wisdom states that no internal organics should persist across geological time, and it has always been assumed no original organic material remains associated with dinosaur fossils.

In light of the data supporting an original source for these materials, and despite long standing assumptions, researchers sought a mechanism to explain preservation of these materials. They reasoned that such a mechanism should be naturally occurring in the immediate environment (i.e. bone) and should increase resistance to degradation and microbial attack, sufficient to stabilize these materials before complete degradation could occur.

In other studies, soft tissues and cells recovered from dinosaur bone elements were frequently associated with iron at levels higher than the surrounding sediments, so researchers hypothesized that iron-mediated reactions (i.e., Fenton-type reactions) played a central role in stabilizing these tissues more rapidly than they

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The Evolution of Understanding

Previous research findings that identified proteins in dinosaur soft tissue seemed to confirm that the tissue was fossilized dinosaur tissue. But there were still doubts in the scientific community about these findings in part because of a lack of understanding about the chemical processes behind such preservation.

Recent research based on ALS studies now shows that the presence of hemoglobin—the iron-containing molecule that transports oxygen in red blood cells—may be the key to preserved ancient proteins within fossils. Hemoglobin is the key—more specifically, the iron contained within the hemoglobin and myoglobin, which is released from its protein cage as the animal begins to decay. Once released, it is highly reactive; in the protein, it is not. Dinosaur blood cells were likely to have more hemoglobin per cell than mammals, which would amplify iron's preservative effect on the tissues.

At the ALS, researchers identified iron particles associated with soft tissues recovered from two Mesozoic dinosaurs. Iron chelators increased fossil tissue immunoreactivity to multiple antibodies dramatically, suggesting a role for iron in both preserving and masking proteins in fossil tissues.

could degrade. Iron is abundant in living cells and tissues, stabilized in the form of heme functional units (as in hemoglobin, myoglobin, or cytochromes), or in the carrier molecule ferritin.

Researchers used transmission electron microscopy, electron energy loss spectroscopy, micro x-ray diffraction, and Fe micro x-ray absorption near-edge structure capabilities at the ALS to characterize the iron associated with fossil tissues, which occurred primarily as the mineral goethite. They then employed experiments to show that iron, derived from hemoglobin lysate, associated with vessels obtained from surviving ostrich bone, and that incubating bone-derived ostrich blood vessels greatly stabilized these otherwise labile materials against microbial attack and degradation. Synchrotron microprobe techniques were used to compare the iron observed in existing hemoglobin-soaked ostrich vessels with iron associated with dinosaur vessels, and showed similar, but not identical, iron moieties. The chemical speciation of iron in the ostrich tissue was a combination of oxyhaemoglobin and a disordered Fe oxyhydroxide that was referred to as a 'biogenic-like oxide', whereas the dinosaur tissues were found to consist of a combination of crystalline goethite and biogenic iron oxyhydroxide. The researchers hypothesize that these represent points on a continuum of biogenic iron to the geological mineral goethite. This mineral is found encapsulated in molecular 'cages' in living systems.

Understanding where living and extinct organisms fit on the 'tree of life' has been limited to those traits persisting in fossilized hard parts, but identifying iron-facilitated reactions that contribute to preservation will provide a better understanding of the relationships of all organisms, living or extinct, on our planet, and perhaps allow the identification of factors contributing to survival, or extinction.

Positing a naturally occurring mechanism for stabilization of soft tissues has implications beyond paleontology. If iron-mediated reactions are part of a continuum from those that facilitate life processes (e.g. cellular respiration, oxygen transport) to fully mineralized, it may be possible to propose a molecular pathway for reactions causing diseases in humans, and once such a pathway is identified, it may be possible to propose ways to interrupt this process.