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Fossil Focus: Diagnosing dinosaurs

by Jennifer Anné[*1]

Introduction

Palaeopathology is the study of the disease and repair of ancient life — most commonly in bone. First coined for the study of diseases in Egyptian mummies, the term was adopted to cover fossil material in 1917 by the first dinosaur doctor, Roy L. Moodie, but has become popular only in recent decades. It is surprising that the study of palaeopathology in the fossil record took so long to catch on in palaeontology. Part of the problem lies with difficulty in getting hold of specimens or accessing the techniques and equipment needed for sensitive analysis. But even if all those problems have been overcome, diagnosing a fossil pathology beyond a vague description brings its own challenges.

Difficulties with diagnosing

Palaeopathologies may be fairly easy for the trained eye to recognize, but diagnosing them is another story. This is due to the loss of soft tissue and finer internal features over time. In addition, dinosaurs represent an area between the pokilothermic (animals that don't make their own body heat) and endothermic (those that do) condition, two metabolisms that can differ in the way the bone responds to stress. For example, dinosaurs are most closely related to two living groups — the crocodiles, with which they share a (rather distant) common ancestor, and the birds, which evolved from dinosaurs. It is hard to tell whether their immune systems might have been like those of crocodilians — highly efficient, rivalling the hardiest of mammal, and allowing them to survive a wide variety of bacterial infections — or more similar to the weaker immune systems of birds. But to understand the pathological afflictions of dinosaurs, you have to first understand the pathways and processes involved in bone healing and repair in their extant relatives.

Healing process

Bone is made primarily of two materials: collagen, an elastic protein that improves fracture resistance; and the harder mineral calcium phosphate, which provides rigidity. It also comes in three flavours — woven, lamellar and fibrolamellar. Woven bone consists of collagen fibres in a haphazard organization, and is characteristic of fast-growing bone. Lamellar bone consists of oriented layers of collagen fibres and is typical of much slower-growing bone. Fibrolamellar bone combines the two, starting with a quick deposition of woven bone that is later filled in by slower-forming lamellar bone. This allows fibrolamellar bone to be both fast-forming (like woven bone) and structurally strong like lamellar bone.

Bone is unique in that it is the only tissue that does not form scars when damaged. This means that during healing, bone must undergo the same processes as it does during development and remodelling. Bone maintenance and repair is therefore an extremely complex process. In fracture healing, there are three main stages (Fig. 1):

Step 1) Inflammation or reactive stage: This occurs immediately after the trauma and is marked by swelling of granulation tissue (connective tissue and blood vessels) surrounding the fracture area. Inflammatory cells enter the area to fight infection, begin clotting and secrete proteins called

cytokines and growth factors. These recruit more inflammatory cells and induce the migration of stem cells that will eventually become important cells such as fibroblasts (which make collagen), chondrocytes (which make cartilage) and osteoblasts (which make bone).

Step 2) Repair stage: The callus begins to form, building up a bridge of connective tissue (collagen) followed by a fibrocartilage plug (a connective tissue made of a mixture of fibrous and cartilaginous tissues) between the fractured areas. This serves as a template for bone formation. Woven bone produced by osteoblasts forms the hard callus, and the removal of cartilage is led by matrix metalloproteinases. These zinc-dependent proteins are also essential for the formation of new blood vessels (angiogenesis) necessary for the ossification process (the formation of new bone).

Step 3) Remodelling: The remaining cartilage is replaced by bone; vascular channels penetrate the woven bone; and osteoblasts form new lamellar bone on the exposed woven-bone surface. In this stage, bone is resorbed by cells called osteoclasts. The osteoclasts (bone-resorbing cells) and osteoblasts (bone-making cells) regulate one another through a series of signal pathways. This feedback maintains the balance of bone resorption and deposition. Eventually, all of the woven bone and cartilage is replaced by mature lamellar bone, thus restoring most of the bone's original strength.

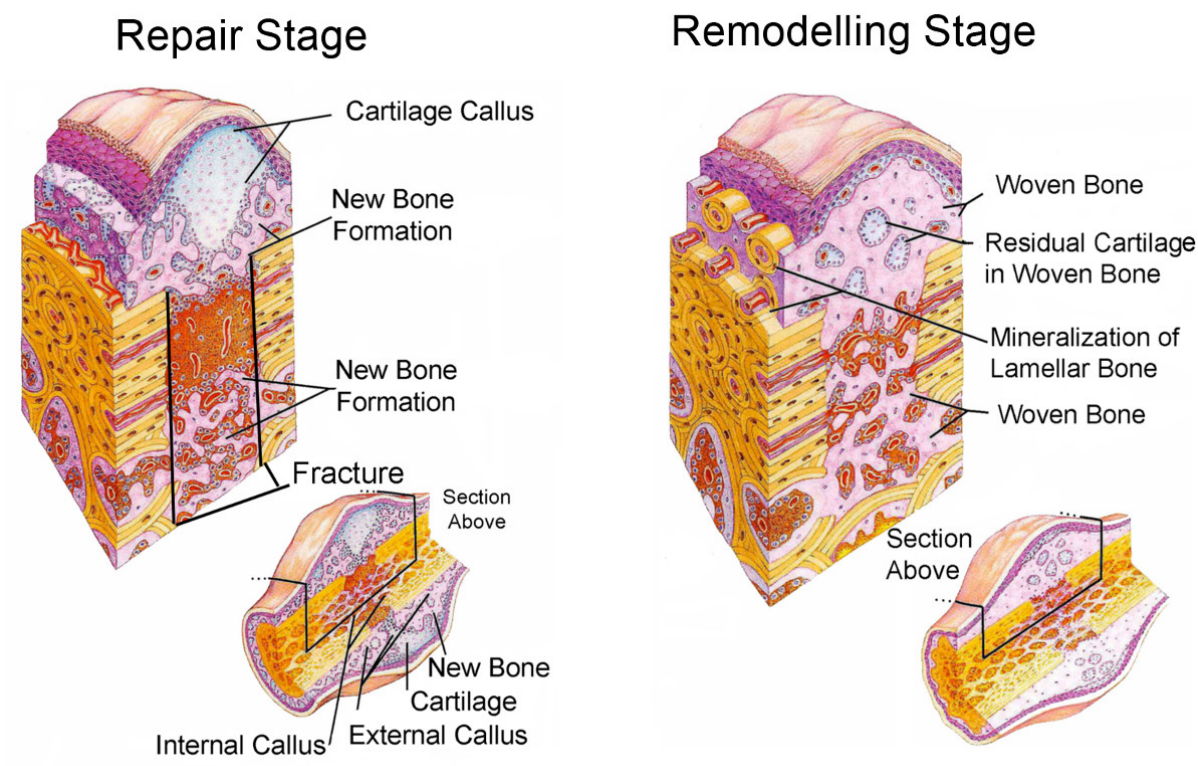

Figure 1 — Diagrammatic representation of the repair (left) and remodelling (right) stages of fracture healing. Left: Bridging of the fracture begins with a cartilage callus and the formation of new bone. Right: Replacement of the remaining cartilage and woven bone by lamellar bone. (Adapted from Ovalle, W. and Nahirney, P. 2008. Netter's Essential Histology (1st Ed.). Saunders Elservier. ISBN 1929007868)

Differences in healing: Are you pokilothermic or endothermic?

Although the general pathways and processes responsible for bone healing and repair are the same in all vertebrates, there are some fine differences between vertebrate groups, such as the rate at which fractures heal and the timing and length of the different healing stages. Reptiles typically have a slower rate of healing than birds and mammals because they generally have slower metabolic rates. Clinically, it can take 6–18 months for a fracture to heal completely in reptiles such as monitor lizards. However, if conditions are favourable (steady supply of food, good temperature and so on), some reptiles can heal remarkably fast, at rates comparable to those recorded in some mammal

species. Specifics of fracture repair (such as healing rate) are poorly understood in birds, which descended from dinosaurs. Birds have been shown to heal faster than mammals in a clinical setting, with a stable fracture taking approximately 3–5 weeks to heal, depending on the injury. However, the duration of fracture healing in wild birds is unknown.

Metabolic differences can also cause differences in bone growth, and thus may be responsible for differences in callus architecture. Pokilotherms (cold-blooded animals) have been shown to form more cartilage and larger calluses than mammals (such as rats), resulting in greater stability in the early stages of fracture healing. This may be because the pokilotherms have a more limited blood supply to the callus than warm-blooded animals. The other extreme can be found in birds, which have been shown to have minimal callus formation in stable fractures.

Getting symptoms from stones

Once you know what to look for, a pathological feature sticks out like, well, a sore thumb. The first step is seeing 'which one of these things is not like the other'. Our eye is good at identifying when something is out of the ordinary: in the case of a skeleton, usually an abnormal texture or shape — a bulge in the middle of a straight bone, a rough texture out of place, and so on. Usually this is enough to identify pathologies. However, there are some animals with bizarre skeletal adaptations that may look pathological at first glance. For example, the skull of a rabbit (*Oryctolagus cuniculus*) has a honeycomb texture that looks like some sort of degenerative bone disease (Fig. 2).

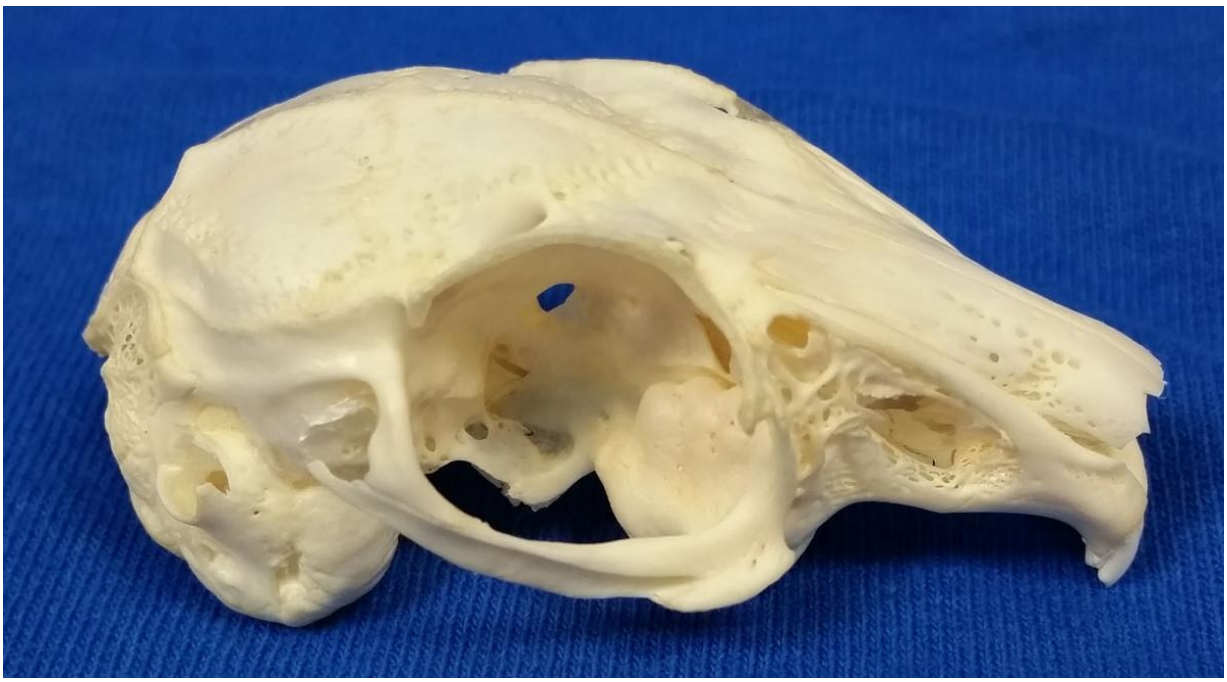

Figure 2 — Rabbit skull, showing the honeycomb texture of the skull. This is normal for Lagomorphs (hares and rabbits). Credit: Timshel Purdum.

In reality, the moth-eaten appearance of the bone is perfectly normal and makes the skull lighter while maintaining strength. The trick to spotting such adaptations is to look for symmetry. Vertebrates (and most other animals) are bilaterally symmetrical, so the left and right should be mirror images of each other. When it comes to dinosaurs, a mounted *Allosaurus fragilis* fossil at the Smithsonian National Museum of Natural History in Washington DC is a good example (Fig. 3).

FIGURE 3 — LEFT (PATHOLOGIC) AND RIGHT (NORMAL) SCAPULA (SHOULDER BLADE) FROM THE MOUNTED *ALLOSAURUS FRAGILIS* AT THE SMITHSONIAN NATIONAL MUSEUM OF NATURAL HISTORY. THE LEFT SCAPULA IS SPLIT IN TWO, PERHAPS BECAUSE OF AN INFECTION, WITH A SMALL BONE SPUR PROTRUDING FROM THE LEFT SIDE.

Sometimes the pathology is not obvious from the outside, especially if it had been healing for some time before the creature died. In these cases, we look to the bone's internal structures and tissue types. For instance, pathological bone usually consists of woven tissue in various stages of compactness. Pathological bone growth also tends to have tissue that is oriented differently to that of the normal bone (Fig. 4).

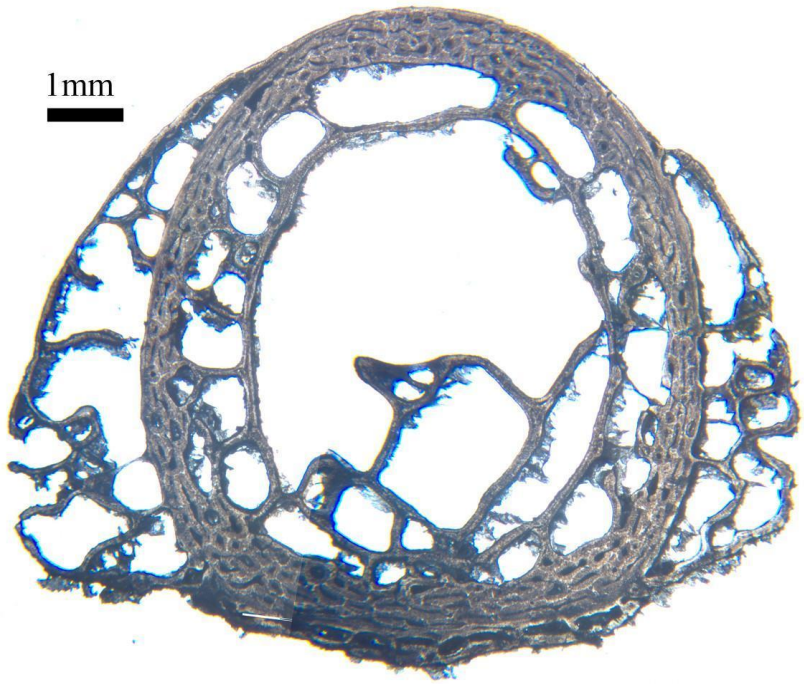

FIGURE 4 — THIN SECTION OF AN ANDEAN CONDOR (*VULTUR GRYPHUS*) TOE BONE (PEDAL PHALANX) UNDER PLANE POLARIZED LIGHT WITH IRREGULAR, PATHOLOGICAL BONE GROWTH. THE PATHOLOGICAL GROWTH CAN BE SEEN AS HAPHAZARD OUTGROWTHS OF BONE RADIATING FROM BOTH THE OUTER AND INNER SURFACES.

Some bone pathologies are degenerative. In these cases, bone resorption is seen as a hole or gap cutting through the normal bone grain, rather than following it (Fig. 5).

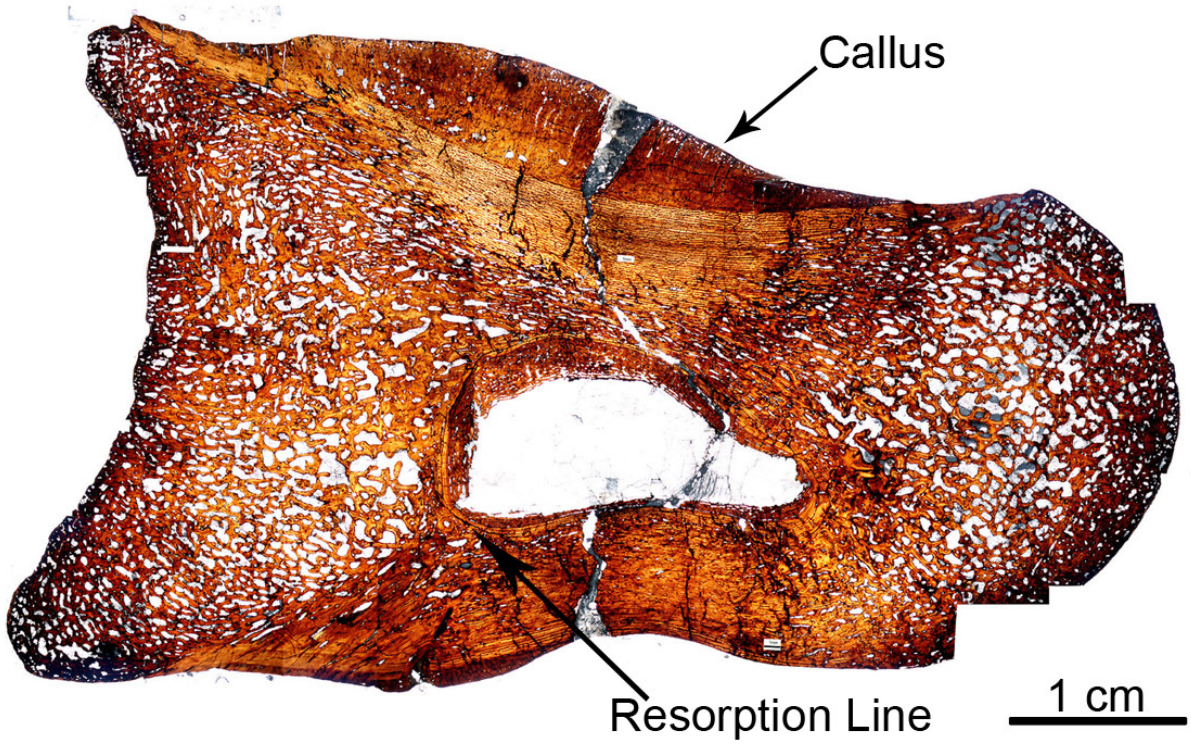

FIGURE 5 — THIN SECTION OF AN *ALLOSAURUS FRAGILIS* (UMNH 6282) PEDAL PHALANX UNDER PLANE POLARIZED LIGHT WITH A CALLUS ON THE TOP SURFACE AND A MEDULLARY CAVITY (CENTRE) SHOWING THE LINE OF RESORPTION.

Diagnosing in the twenty-first century

The past few years have allowed us to develop new tools for identifying and diagnosing palaeopathologies. In particular, we can now map and characterize the trace elements crucial for bone repair. There are a number of different biosynthetic pathways involved in bone repair and remodelling, and many of these pathways are aided (or deterred) by specific trace elements, mainly copper, zinc and strontium.

Copper is important for collagen, the main organic constituent of bone. It creates a molecular cross-link that prevents the collagen from unravelling. Zinc is important for a number of biosynthetic pathways in bone, and is associated with areas of active bone formation. Strontium has been found to increase bone deposition by interrupting one of the pathways by which bone is resorbed, and has even been pursued as a drug (called strontium ranelate) to treat osteoporosis . Levels of these trace metals are elevated around an infected area during the distinct stages of fracture healing, bone development and remodelling (Fig. 6). As a result, looking at the distribution, concentration and coordination of these elements in bone now allows us to identify biosynthetic processes in bone repair occurring up to the time of death.

Further reading

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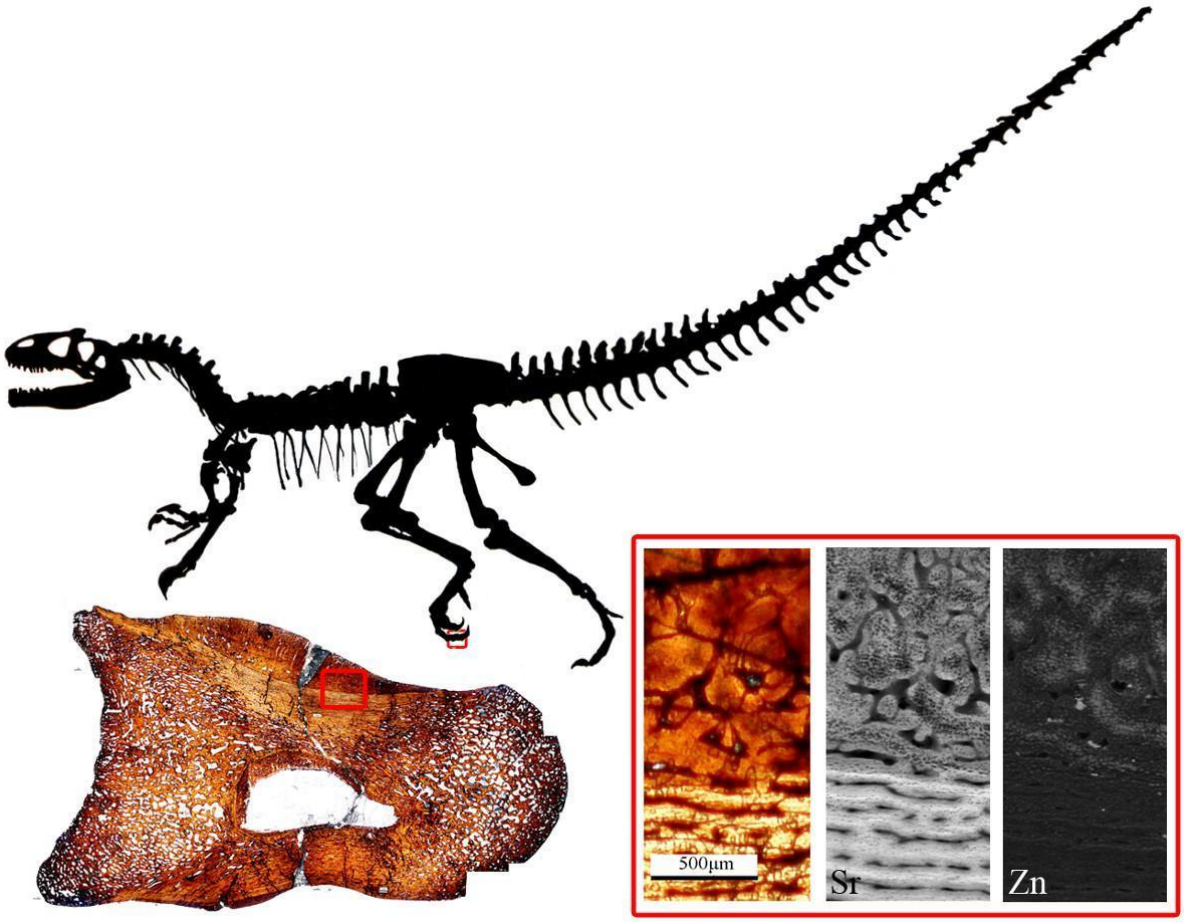

FIGURE 6 — THE SAME *A. FRAGILIS* TOE AS IN FIG. 6, WITH ITS LOCATION MARKED ON THE ALLOSAURUS OUTLINE; A CROSS SECTION OF THE FOSSIL; AND THE CHEMISTRY OF THE CALLUS/NORMAL-BONE INTERFACE REVEALED BY SYNCHROTRON ELEMENTAL MAPPING (MICROFOCUS BEAMLINE I18, DIAMOND LIGHT SOURCE). IN THE CALLUS, STRONTIUM (SR) SHOWS DISCRETE TISSUE FEATURES NOT OBSERVABLE IN CONVENTIONAL THIN SECTION, ALLOWING FOR A BETTER ESTIMATE OF HOW MUCH HEALING HAS OCCURRED. ADDITIONALLY, ZINC (ZN) IS ELEVATED IN THE WOVEN TISSUE — THE HEALING PART — OF THE CALLUS COMPARED TO THE NORMAL BONE. IN MODERN ANIMALS, ELEVATED ZINC IN BONE IS ASSOCIATED WITH AREAS OF ACTIVE OSSIFICATION, SUCH AS IN REMODELLING CALLUSES.

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