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Article

A flattened enantiornithine in mid-Cretaceous Burmese amber: Morphology and preservation

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ABSTRACT

Cretaceous amber from Myanmar (~99 Ma Burmese amber) has become a valuable supplement to the traditional skeletal record of small theropod dinosaurs preserved in sedimentary rocks, particularly for coelurosaurs and enantiornithines. The specimens recovered from this deposit preserve skeletal material and soft tissues in unmatched detail. This provides opportunities to study three-dimensional preservation of soft tissues, microstructure, and pigmentation patterns that are seldom available elsewhere in the fossil record. Ultimately, this line of research provides insights into life stages that are difficult to preserve, the ecology and appearance of the groups involved, and the evolutionary-development of structures such as feathers. Here we describe the most recent discovery from Burmese amber, an articulated skeleton of an enantiornithine bird. This individual has been sectioned along the coronal plane, providing a unique view inside multiple body regions. Osteological observations and plumage patterns support placement within the Enantiornithes, and suggest that the animal may have been a juvenile at the time of death. The specimen has a complex taphonomic history that includes exposure at the surface of a resin flow prior to encapsulation, and may include scavenging by some of the insects trapped within the same amber piece. The chemical composition observed along surface exposures and shallowly buried regions of the body indicate that the specimen has not undergone significant exchange with its surroundings. High iron concentrations are present in regions that preserve soft tissues as carbon films, and calcium distribution corresponds to regions where bones breach the surface of the amber.

1. Introduction

Recent discoveries of mid-Cretaceous amber from Myanmar (reviewed by [1–3]) have provided unprecedented information for understanding the early post-natal development of early birds. Over the last two years, Xing et al. [4–6] have described two precocial Enantiornithes wings and a partial hatchling, as well as a feathered coelurosaurian tail with primitive plumage from mid-Cretaceous Burmese amber. These new findings associate well-preserved feathers with skeletal material for the first time. They also highlight the unique preservation potential of amber for understanding the morphology and evolution of vertebrate integumentary structures. However, despite these advances, findings over the last two years have largely been limited to partial or appendicular skeletal material, due to the small sizes of the amber pieces involved.

To date, enantiornithines remains in amber consist of one isolated wing tip (DIP (Dexu Institute of Palaeontology, Chaozhou City, China)-V-15101), that may have been disassociated from the corpse through decay or predation; one wing tip

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(DIP-V-15100) that appears to have been part of a much more complete skeleton, which was destroyed during the mining process or subsequent polishing of the surrounding amber in preparation for jewelry manufacture [4]; and a partial hatchling (HPG (Hupoge Amber Museum, Tengchong City Amber Association, China)-15-1) that included most of the skull, part of the neck, one wing, and both feet [6]. The size bias that is prevalent in the amber fossil record [7] has meant that most of the finds in Burmese amber have belonged to small, precocial juveniles the size of modern hummingbirds. These specimens have supplemented our understanding of Enantiornithes compression fossils with comparatively limited soft tissue preservation in Cretaceous rocks from Argentina [8], Brazil [9], China [10–12] Mongolia [13], Spain [14,15], and the USA [16].

Preservation among the Burmese amber skeletal specimens has been variable, depending on the size of the specimen and the degree to which the remains have been exposed to weathering, decay, and compaction prior to full resin polymerization. In the best-preserved specimens, bones are found in articulation and without significant deformation or replacement (e.g., DIP-V-15100, where intact osteon structures are visible; [4]). However, deformation of the surrounding resin mass has shattered, scattered, or compressed the thin and hollow bones of some specimens (e.g., HPG-15-1, where the metatarsals and digits are encapsulated in soft tissue, but the bones are fragmentary; [6]). Where bones have been exposed at the amber surface, providing a conduit for pore waters, some specimens exhibit infilling or partial replacement of bones by clay minerals (e.g., DIP-V-15103, where bone replacement limits X-ray μCT scanning contrast; [5, Fig. 1]).

The areas available for chemically examining preservation within the amber have been limited by the available surface exposures, but SR μXFI (synchrotron radiation micro-X-ray fluorescence imaging) and XAS (X-ray absorption spectroscopy) have proven valuable as non-destructive tools for mapping the chemistry and probing the oxidation state of individual elements within these amber specimens [5]. In combination, these techniques have shed additional light on the preservation process for vertebrate inclusions within Burmese amber, permitting comparisons to discoveries in sedimentary rocks (e.g., [16–18]).

Here we describe the contents of the largest piece of Burmese amber with theropod inclusions known to date: an enantiornithine including much more of the axial skeleton. This specimen allows us to examine preservational processes in greater detail, providing the first internal view of multiple body regions in one of these inclusions, and an expansive surface for chemical mapping.

2. Materials and methods

2.1. Specimen and photography

The new amber specimen, DIP-V-15102, comes from the Angbamo site, Tanai Township (Myitkyina District, Kachin Province) of northern Myanmar (Fig. 1). It measures approximately 68 mm × 7 mm × 51 mm, and weights 14.99 g. The original specimen is housed and displayed in the Dexu Institute of Palaeontology (=DIP), China.

DIP-V-15102 was examined using a Leica MZ 12.5 stereomicroscope with a drawing tube attachment. Photographs were taken using a Canon digital camera (5D Mark III, MP-E 65MM F/2.8 1–5X) fitted to a macro rail (Cognisys), and were processed using Helicon Focus 5.1 and Adobe Photoshop CS5 software to increase depth of field in the images. These images were supplemented with photos taken under long wavelength UV light (395 nm), mapping resin flows.

2.2. Micro-CT scanning and 3D reconstruction

DIP-V-15102 was scanned with a MicroXCT 400 (Carl Zeiss X-ray Microscopy, Inc., Pleasanton, USA) at the Institute of Zoology, Chinese Academy of Sciences. The scans of the entire animal were completed by dividing the specimen into seven scans that were done with a beam strength of 60 kV, 8 W, absorption contrast and a spatial resolution of 43.3249 μm. The different parts of the bird (head, hip, apical forelimb, radius-ulna area, dorsal vertebrae) were scanned separately in higher resolution, with a beam strength of 60 kV, 8 W, absorption contrast and the spatial resolution of 19.4520 μm, 20.9483 μm, 20.9483 μm, 4.2520 μm, 4.2520 μm, respectively.

Based on the image stacks obtained, structures of the specimen were reconstructed and separated with Amira 5.4 (Visage Imaging, San Diego, USA). The subsequent volume rendering and animation were performed with VG Studiomax 2.1 (Volume Graphics, Heidelberg, Germany). Final figures were prepared with Photoshop CS5 (Adobe, San Jose, USA) and Illustrator CS5 (Adobe, San Jose, USA).

2.3. μ-XFI chemical mapping

The μ-XFI analysis was performed at 4W1B beamline, Beijing Synchrotron Radiation Facility, which runs 2.5 GeV with a current of 250 mA in SR-dedicated mode. The incident X-ray is monochromatized by the W/B4C Double Multilayer Monochromator at 15 keV and is focused down to 100 μm in diameter by a polycapillary lens. The two-dimensional mapping is acquired in step-mode: the sample is held on a precision motor-driven stage, scanning 100 μm stepwise. The Si(Li) solid state detector is used to detect X-ray fluorescence emission lines with an exposure time of 20 s. Data reduction and processing were performed using the PyMca software package [19]. Herein, the feather and skin terminology presented by Lucas and Stettenheim [20] is largely followed, while details related to barbule morphology and pigmentation follow Dove [20].

3. Results

3.1. Osteological characters

DIP-V-15102 is an articulated skeleton (Fig. 2). The dense feathers and their well-developed rachises suggest this specimen represents a bird. Most avian records in Burmese amber consist of isolated feathers or partial bones. Consequently, the discovery of an articulated skeleton is an extremely rare event. DIP-V-15102 was cut and polished by a local miner along the coronal plane prior to study, losing the rostral and middle portions of the skull and most of its right wing and leg. However, the surviving skeletal portions still render the specimen the most complete individual discovered thus far in Burmese amber.

Remains of the right basicranium (part of the frontal and parietal region), axial column (about 5 cervical vertebrae and 8 dorsal vertebrae), forelimb (distal right humerus, radius and ulna), partial pelvic girdle and right femur are preserved in articulation (Fig. 2c, d). All bones are very compressed and the degree of co-fusion between elements is obscured by the quality of the scan. The cervical vertebrae preserve no clear anatomical information. The thoracic series consists of nine to ten vertebrae in articulation, displaying large square-shaped neural spines. The sacral vertebrae can be somewhat distinguished suggesting that fusion of the synsacrum was incomplete. A few free caudal vertebrae can be discerned. They appear to be followed by a robust triangular element interpreted as the pygostyle. The pubes are U-shaped, strongly concave medially, with a short distal symphysis, as in