Supporting Information

Pradel et al. 10.1073/pnas.0807047106

SI Text

Methods of X-Ray CT. The specimen OKM38 has been scanned at the University of Texas High-Resolution X-ray CT Facility, Austin. Scan parameters were as follows: 1,024 × 1,024 16-bit TIFF images. II, 180 kV, 0.12 mA, no filter, air wedge, no offset, slice thickness 2 lines (0.04872 mm), S.O.D. 70 mm, 1,400 views, 2 samples per view, interslice spacing 2 lines (0.04872 mm), field of reconstruction 22.8 mm (maximum field of view 23.17 mm), reconstruction offset 8,000, reconstruction scale 3,200, acquired with 19 slices per rotation and 15 slices per set. Flash and ring-removal processing was done based on correction of raw sinogram data by Alison Mote by using IDL routines "RK_SinoDeSpike" and "RK_SinoRingProcSimul," both with default parameters. The first 4 duplicate slices of each rotation, except for slices 1–4, were deleted. Rotation correction processing was done by using IDL routine "DoRotationCorrection." Final slices totaled 649.

Virtual Reconstruction. To complete visualization, segmentation, and 3D rendering (Movie S1 and Movie S2), we have used MIMICS software (Materialise Inc.; proprietary software at the Museum National d'Histoire Naturelle, Paris). MIMICS (Materialise's Interactive Medical Image Control System) is of great interest to deal with such a large number of datasets as those generated from synchrotron microtomography and holotomography. We have used the MIMICS 64-bit version running on a Dell 690 Windows XP 64 workstation with 16 GB of RAM.

MIMICS allows different types of measurement and segmentation to be performed. Regions of interest can be selected with accuracy by using the threshold method to create segmentation masks. With this method, selections depend on a range of defined gray values and not on manual outlining operations. Three-dimensional models have been calculated from segmentation masks and combined through Boolean operations.

Microprobe Analysis. We have carried out microprobe analysis on the specimen KUNHM 22060 at the level of the only areas where the structure referred to as the "brain" and "cranial nerves" reaches the surface of the specimen; that is, at the level of the right optic nerve foramen (Fig. S1 *A* and *B*; the natural cast of the orbital cavity was separated from the rest of the nodule on the right side) and of an almost transverse break through the nodule, which shows the calcite filling of the brain cavity and possibly the rearmost end of the presumably preserved spinal cord. In both cases, SEM images show a crystalline structure that radically differs from that of the surrounding calcite and proved to be composed of calcium phosphate (Fig. S1 *C–E*).

The analysis was operated by Omar Boudama at the Electron Microscopy Service of the UFR 928, University Paris 6, by using a field effect gun (FEG) scanning electron microscope Zeiss SUPRA55VP, with a nominal resolution of 1.0 nm (at 15 kV and 2 mm distance). The analyzer was a detector SDD SAHARA of PGT including the SPIRIT software.

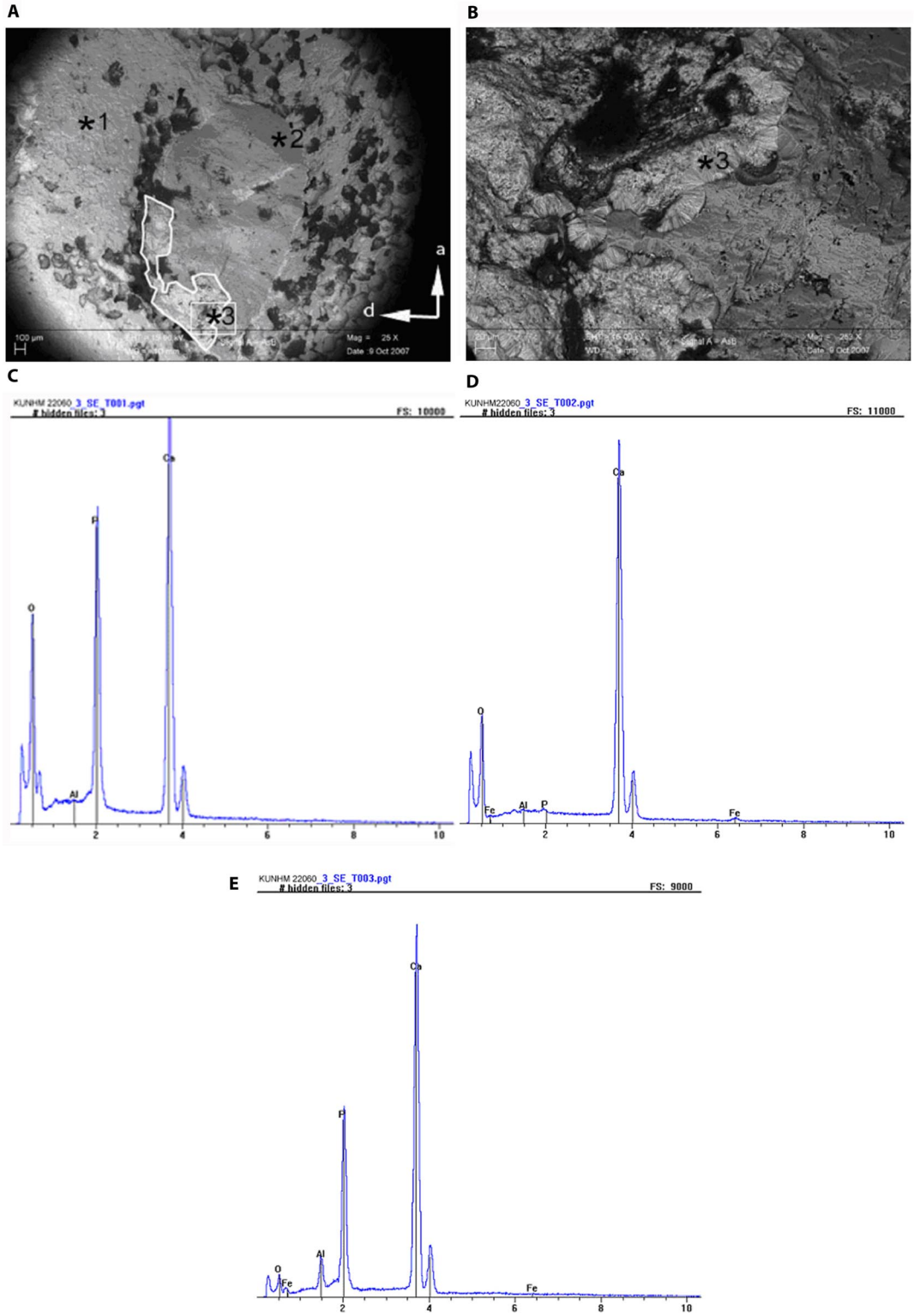
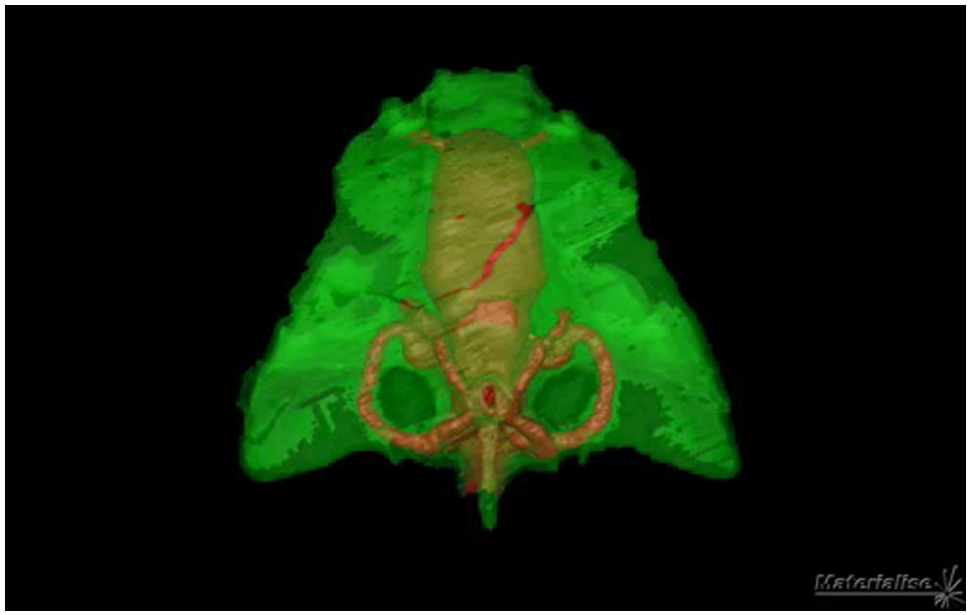
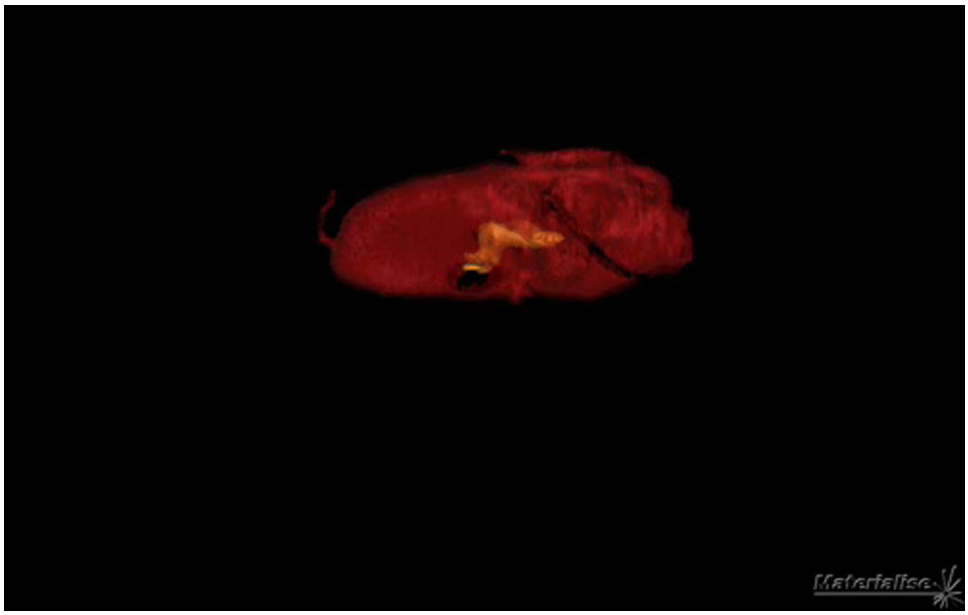


Fig. S1. KUNHM 22060, SEM micrograph and microprobe analyses of the surface of the calcite-filled right optic foramen. The asterisks in *A* and *B* indicate the location of the microprobes analyses corresponding to the respective diagrams in *C–E*. (*A*) General view of the optic foramen and surrounding prismatic calcified cartilage; arrows point anteriorly (a) and dorsally (d); the area delimited by a bold line corresponds to the emergence of the preserved optic nerve; calcium phosphate dominates in the light-gray areas, and calcium carbonate dominates in the dark-gray areas. (*B*) Detail of the area framed in *A*, showing the finely crystalline structure of calcium phosphate of the preserved optic nerve. (*C–E*) Microprobe analyses for P, O, Al, Fe, and Ca at the level of the optic foramen and ''optic nerve'' of KUNHM 22060 (*1–*3 in *A* and *B*). (*C*) External surface of the prismatic calcified cartilage (*1, mostly calcium phosphate). (*D*) Surface of the calcite filling the brain cavity and cranial nerve canals (*2, calcium carbonate). (*E*) Surface of the emerging preserved optic nerve (*3, mostly calcium phosphate with traces of Al and Fe).



Movie S1. Braincase (green), brain cavity and labyrinth cavity (red), and presumed phosphatized brain (yellow) of the Pennsylvanian sibyrhynchid KUNHM 22060, rotating from dorsal to ventral view. Three-dimensional virtual reconstruction made from SR-μCT images by means of MIMICS (Materialise, Inc.).

Movie S1 (MOV)



Movie S2. Braincase (green), brain cavity and labyrinth cavity (red), and presumed phosphatized brain (yellow) of the Pennsylvanian sibyrhynchid KUNHM 22060, rotating from anterior to posterior view. Three-dimensional virtual reconstruction made from SR-μCT images by means of MIMICS (Materialise, Inc.).

Movie S2 (MOV)