IDENTIFICATION OF CHITIN PRESERVED IN A PENNSYLVANIAN AGE FOSSIL SCORPION CUTICLE. N. S. Gupta1, A. L. D. Kilcoyne2, D. E. G. Briggs3, R. E. Summons4, and G. D. Cody5, 1Geophysical Laboratory, Carnegie Institution of Washington, DC 2Advanced Light Source, Lawrence Berkeley Laboratory, CA, 3Dept. of Geology and Geophysics, Yale University, CT, 4Dept. of Earth, Atmospheric, and Planetary Sciences, Massachusetts Institute of Technology, MA

Introduction: The organic fossil record of chitinous organisms including fossil arthropods (insects, scorpions, eurypterids), cephalopods and decapods spans back to the early Cambrian. The exoskeleton of arthropods is a complex laminate, where different layers serve different biological functions. The most chemically recalcitrant layers, most likely to persist in the fossil record, are the hard exocuticle, a composite of chitin fibers embedded in a protein matrix, that is cross-linked (sclerotized) by catechol, aspartate and histidyl moieties, and the epicuticle, a waxy layer that protects the arthropod from desiccation. The conventional geochemical view would not predict that chitin (poly-N-acetyl-D-glucosamine) would be preserved in ancient fossils. Indeed, the oldest confirmed chitin has been identified in a ~ 22 my old, remarkably well preserved, fossil cuticle [1]. Analysis of older samples using pyrolysis GC-MS revealed no evidence for chitin in a wide range of Paleozoic arthropod fossils[2].

The development of high-brilliance synchrotron X-ray sources, coupled with advances in soft X-ray optics (soft X-rays being defined as spanning the energy range ~ 200 to 1200 eV), has led to the development of Synchrotron-based Scanning Transmission X-ray Microscopes (STXMs). In the present study, carbon, nitrogen, and oxygen micro X-ray Absorption Near Edge Structure (C-, N-, and O-µXANES) spectromicroscopy is applied to the analysis of fossil arthropod cuticle to provide in-situ molecular spectroscopic information at sub-micron (i.e. down to 25 nm) scales. The combination of high spatial and energy resolution yields C-, N-, and O-XANES spectra from sub-micron scale domains and provides an accurate assessment of the types of organic functionality present and, in the case of fossil organics, allows us to ascertain the biomolecular assemblage preserved.

Results: Analysis of modern scorpion cuticle reveals the expected and considerable compositional variation across the various laminae. For example, in fig. 1, we present a nitrogen K edge map obtained by acquiring two high resolution images, one at 395 eV (just below the N-K edge) the other at 410 eV (on top of the N-K edge). The –log (Iy/I1) image provides a “map” of the distribution of nitrogen where nitrogen rich regions appear as light regions. In fig. 2 we perform the same analysis on a fossil scorpion (Pennsylvanian in age, ~ 320 my old). In addition, we have performed high resolution C-, N-, and O-XANES spectra on the various regions from both scorpion cuticles. These data clearly reveal the presence of considerable nitrogen, with N-organic functionality that is consistent with the preservation of at least some chitin in the fossil. A complete functional group and elemental analysis provides a consistent chemical explanation as to how chitin is preserved over such an extensive period of Earth’s history.

Figure 1: Nitrogen K-edge “Map” through a cross section of a modern scorpion cuticle where lighter regions are richer in nitrogen. Note the scale bar is 5 μm.

Figure 2: Nitrogen K-edge Map image across 320 my old scorpion cuticle. Light regions reveal high concentrations of nitrogen and preservation of the laminar compositional variation. Scale bar is 1 μm.