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Preservation of Protein in Phytodetritus and Sediments via Macromolecular Aggregations

Reno Nguyen & **Rodger Harvey**

Chesapeake Biological Lab/UMCES, Box 38, Solomons, MD, 20688-0038, USA

The abundance of proteins in living organisms has sparked considerable interest into the study of their fates in the aquatic environment. Historically, proteins have been considered very labile in the environment, and consequently unlikely to survive as high molecular weight components during early diagenesis. Evidence is increasing, however, that some fraction of proteinaceous material is preserved in freshwater, estuarine, and marine environments and our previous work demonstrated that up to 60% of residual nitrogen in algal detritus and in sediments that can be attributed to high molecular weight (>2 kDa) proteinaceous material (Nguyen and Harvey, 1997). Here we report the fate of proteins during early diagenesis in environments with low mineral content to assess their preservation via mechanisms other than mineral sorption. Preservation was determined for anoxic, organic-rich sediments of Mangrove Lake, a marine environment located in Bermuda, and for algal detritus comprised principally of diatomaceous material generated during oxic decay experiments. N-phenacylthiazolium bromide (PTB) was used test the hypothesis that proteins may undergo modification reactions with glucose and other reducing sugars to form a particular class of products termed advanced-glycation end-products (AGEs). A small but significant release (14% more) of proteins was observed with PTB in surficial sediments, but other covalent cross-linkages, appear to be important in the older sediment sequences. Size-exclusion HPLC with protein fluorescence, absorbance, and evaporative light scattering detector measurements under native (phosphate or bicarbonate buffers) and denaturing (guanidine HCl, urea, or acetonitrile) conditions point to macromolecular aggregations by strong hydrophobic interactions as a major mechanism for the preservation of proteinaceous material. These soluble aggregates of substantial molecular weight (1.5 kDa) appear to be formed early in the diagenetic sequence. The preferential preservation of aggregated or high molecular weight, multi-subunit phytoplankton proteins in sediments suggests that such aggregations confer resistance to enzymatic attack. Soluble aggregates comprised most or all of the acid hydrolyzable proteinaceous material in detritus and surficial sediments but <35% in 9.7 m deep sediments. Buffer-extractable material in the deepest sediments were significantly enriched in glycine compared to surficial sediments (24 vs. 14 mole%), indicating the hydrophobic nature of the residual material and the possible contribution of bacterial protein. Extended (18 h) incubations with trypsin covalently linked to agarose beads showed that much of the aggregates in the diagenetically processed samples, are susceptible to proteolytic cleavage. These results are direct evidence for the preservation of peptide linkages in sediments as old as 4000 years and for hydrophobic associations of protein as an important mechanism in long-term preservation.

Nguyen RT & Harvey HR, *ACS Symposium Series*, **707**, 88-112, (1998).