Materials Nanoarchitecturing via Cation-Mediated Protein Assembly: Making Limpet Teeth without Mineral

Abstract

Teeth are designed to deliver high forces while withstanding the generated stresses. Aside from isolated mineral-free exception (e.g., marine polychaetes and squids), minerals are thought to be indispensable for tooth-hardening and durability. Here, the unmineralized teeth of the giant keyhole limpet (Megathura crenulata) are shown to attain a stiffness, which is twofold higher than any known organic biogenic structures. In these teeth, protein and chitin fibers establish a stiff compact outer shell enclosing a less compact core. The stiffness and its gradients emerge from a concerted interaction across multiple length-scales: packing of hydrophobic proteins and folding into secondary structures mediated by Ca2+ and Mg2+ together with a strong spatial control in the local fiber orientation. These results integrating nanoindentation, acoustic microscopy, and finite-element modeling for probing the tooth's mechanical properties, spatially resolved small- and wide-angle X-ray scattering for probing the material ordering on the micrometer scale, and energy-dispersive X-ray scattering combined with confocal Raman microscopy to study structural features on the molecular scale, reveal a nanocomposite structure hierarchically assembled to form a versatile damage-tolerant protein-based tooth, with a stiffness similar to mineralized mammalian bone, but without any mineral.

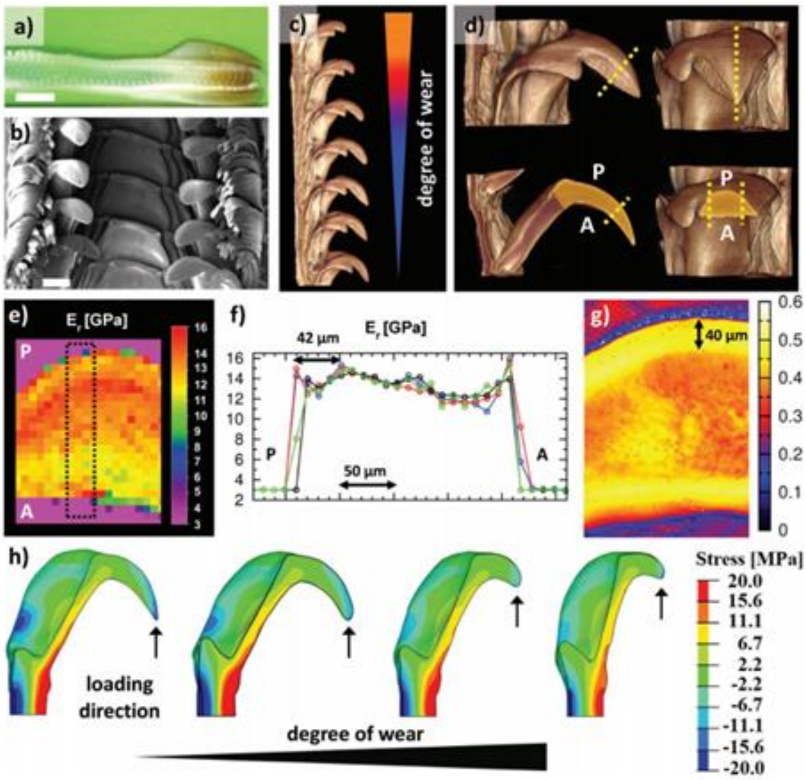

Figure 1. Morphology, wear, and mechanics of teeth. a) An optical image (scale bar = 5 mm) and b) Scanning-electron microscopy (SEM) image of the *M. crenulata* radula (scale bar = 500 µm); c) Micro-computed tomography (µ-CT) reconstruction of an array of working teeth depicting progressive signs of wear; d) magnified µ-CT images of a fully formed and operational tooth from (a) depicting two types of sections used for structural and mechanical analysis, longitudinal (left) and transverse (right). P and A denote the posterior (trailing) and anterior (leading) edges of the tooth. e) Maps of reduced modulus on a transverse section depicted by dashed lines in the left panel of (d); f) superimposed adjacent profiles of reduced modulus measured in the area designated in (e); g) scanning acoustic microscopy image depicting the acoustic reflectivity of an area of a transverse section along the plane depicted in bottom right panel of (d). The dashed lines in panel (d) depict the sectioning planes investigated by mechanical testing shown in (e) and (g). h) FEM simulations of longitudinal stresses generated within teeth as a result of applying a force of 125 mN to the tooth tip in the vertical direction (denoted by the arrow). The degree of wear increases toward the right. No significant differences are observed for different stages of wear, demonstrating a versatile wear-tolerant tooth design. The results for three additional loading directions and the corresponding displacement maps are displayed in Section S3 (Supporting Information). Note that the stresses and deformations can depend strongly on the direction of applied force, and *M. crenulata* teeth are found to be well adapted to different loading scenarios (Section S3, Supporting Information).