Conductivity and dynamics of biomimetic synthetic polypeptides

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Squid-inspired synthetic proteins self-assemble into a hydrogen-bonding physically crosslinked network that gives rise to remarkable thermal and protonic conductivity. Understanding the chain dynamics of biomimetic structural proteins and their role in heat and proton transport will help design novel biodegradable protein-based materials with programmable properties for bioelectronics and thermal switching devices.

Proteins are heteropolymers that provide a variety of building blocks for designing novel biological materials. Proteins are diverse but often display substantial similarity in sequence and three-dimensional structure. A new family of repetitive structural proteins with remarkable properties was recently identified in the tentacles of several squid species. Using the tools of molecular biology and proteomics, we demonstrated that these squid ring teeth (SRT) proteins have segmented semi-crystalline morphology with repetitive amorphous and crystalline domains. However, a clear relationship between the protein architecture (repetition and length of crystalline/amorphous domains), the self-assembled nanostructure, and conducting properties remains elusive due to complexity of native amino acid sequences.

To investigate the genetic basis of material properties in natural and artificial SRT sequences, we have developed a new approach for the design and production of structural proteins. These synthetic proteins have identical sequences but increasing repeat unit number *n* (*n* = 4, 7, 11 and 25) and molecular weights (15, 25, 42 and 86) kDa. Synthetic SRT proteins have remarkable protonic conductivity in the range of (1 to 5) mS/cm, which is the highest reported to date for protein-based materials as measured by Electrochemical Impedance Spectroscopy. In addition, synthetic SRT proteins exhibit a programmable thermal conductivity from 0.3 W/mK to 1.4 W/mK.

To investigate these vibrational dynamics, we turned to elastic and quasi-elastic neutron scattering (QENS) using the High-Flux Backscattering Spectrometer (HFBS) and Disk-chopper time-of-flight spectrometer (DCS) at the NIST Center for Neutron Research. QENS can measure molecular dynamic processes such as rotations, relaxations and diffusive motions with 1 Å to 30 Å and pico- to nanosecond resolution by directly probing

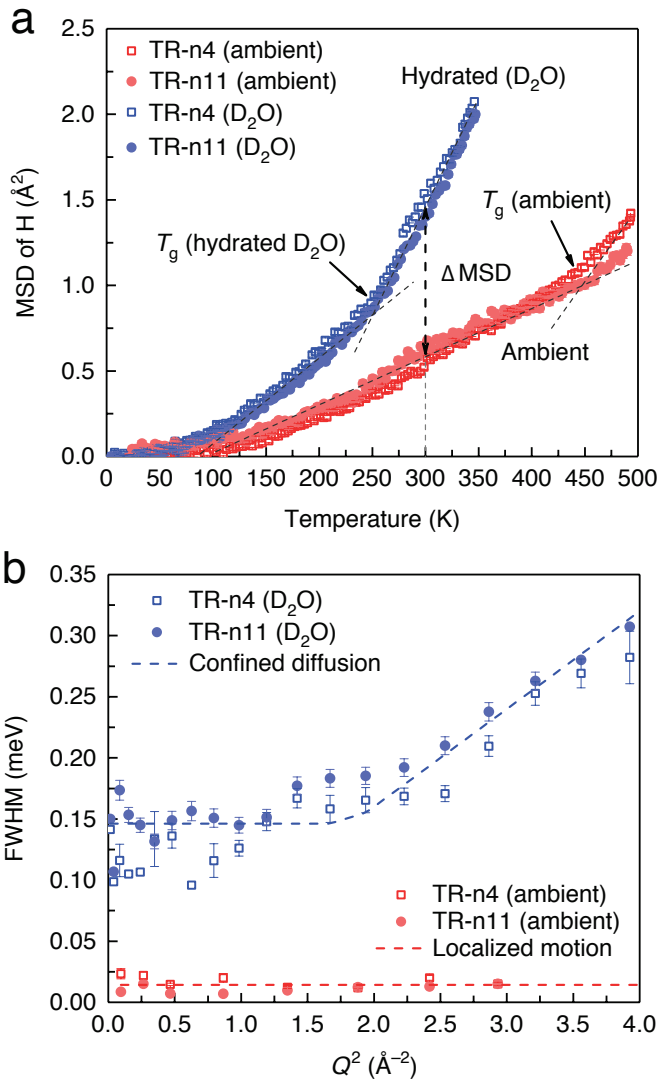

FIGURE 1: a) Measured mean square displacement, or vibrational amplitude, of hydrogen atoms in the TR films as a function of sample temperature. As seen, the dense, hydrogen-bonded network opens up and allows for increased movement following hydration. b) Full-width at half-maximum of the quasi-elastic peaks of the ambient and hydrated proteins as a function of the square of the scattering wavevector, Q^2. Error bars represent one standard deviation.

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the self-diffusion of hydrogen atoms. The hydrated chain dynamics were measured using deuterated water (D_2O), as deuterium has a negligible neutron scattering cross-section and therefore it does not contribute to the signal. The mean square displacement (MSD) of hydrogen atoms in the protein (shown in Fig. 1a) was calculated from the scattering intensity in fixed window elastic scans, as a function of temperature, using the Debye–Waller factor (which is a standard Gaussian approximation method). Tandem Repeat (TR) polypeptides under ambient conditions show very localized motions at $T > 70$ K, which is common for proteins and typically originates from methyl group rotations, and exhibit a glass transition around 450 K. Therefore, neutron spectroscopy does not show significant segmental or backbone motion at room temperature (300 K). Thus, the disordered chains are constrained in a dense hydrogen-bonding network with minimal vibrational freedom. Conversely, in the hydrated state, the MSD of the hydrogen atoms increases and we observe much larger vibrational amplitudes, as well as a decrease in the glass transition temperature (250 K). In this case, the water molecules break the hydrogen bonding between the disordered chains, allowing for more delocalization of the hydrogen motion. Protein chain dynamics were further investigated by quasi-elastic measurements. Figure 1 shows the full-width at half-maximum, $\Gamma(Q)$, of the quasi-elastic peaks of ambient and D_2O-hydrated TR-n4 and TR-n11 proteins, plotted as function of Q^2. Under ambient conditions (<1 % relative humidity during our QENS measurements), TR proteins show a Q-independent behavior (15 μeV) characteristic of localized motions (that is, methyl group rotations). On the other hand, D_2O-hydrated TR proteins show a Q-independent plateau at low Q values (0.15 meV), while a linear scaling with Q^2 is observed at higher Q. This two-regime Q dependence is characteristic of diffusion in a confined space and can be described by the Volino and Dianoux (VD) model for bounded diffusion in a potential of spherical symmetry, which corresponds to confined diffusive motions of the amorphous segments within the β-sheet nanocrystals.

In order to understand the transport processes at the molecular level, we have designed variants of squid-inspired proteins with variable repeat unit number (n). Our results of elastic fixed window scan data collected at HFBS shows the hydration dynamics of two variants of biomimetic SRT proteins changes upon hydration for both polypeptides. QENS analysis at HFBS and DCS are necessary to understand the protein chain dynamics. Based on these results, we concluded that the hydration plays a key role in localized motions, and faster self-diffusion of amorphous chains in SRT polypeptides, which enhances the thermal conductivity. The thermal conductivity values scales with increasing repeat unit number n (enabling precise control of the physical properties by sequence design), and are highly dependent on protein hydration (enabling fast reversible switching of thermal conductivity), with a switch ratio of 4.5 at room temperature, which is among the highest reported to date.