Protein Dynamics from High-Resolution Relaxometry

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A proper understanding of the physics and chemistry that underlie the function of biological macromolecules requires an atomic-resolution description of their conformational space and the timescales of the motions in this space. Methodological and computational developments over the last three decades have made nuclear magnetic resonance a mainstream experimental technique to characterize dynamics of biomolecules, and particularly proteins. The measurement of nuclear spin relaxation has given access to motions on multiple timescales between the tens of picoseconds and nanoseconds. However, the analysis of motions has relied on limited experimental data (most often only three relaxation rates per residue in a protein) and used simple model-free approaches. High-resolution relaxometry, as introduced by Redfield [1], is a powerful technique to quantify nuclear spin relaxation over a broad range of magnetic fields and provides unprecedented sets of experimental data to quantify protein motions in the ps-ns range.

Here, we show that high-resolution relaxometry, combined with high-field relaxation measurements, opens the way to a more accurate description of picosecond to nanosecond motions in proteins. Our first high-resolution relaxometry study of backbone nitrogen-15 on the protein ubiquitin has revealed motions on low-nanosecond time scales [2,3]. Here, we present carbon-13 relaxation rates recorded between 0.3 and 18.8 T on methyl groups in ubiquitin (Fig. 1). The resulting extensive ensemble of experimental constraints allows the analysis of motions with up to three correlation times spanning three orders of magnitude from picoseconds to nanoseconds. Experimental results agree reasonably well to the analysis of a 0.5 µs trajectory calculated by molecular dynamics (MD).

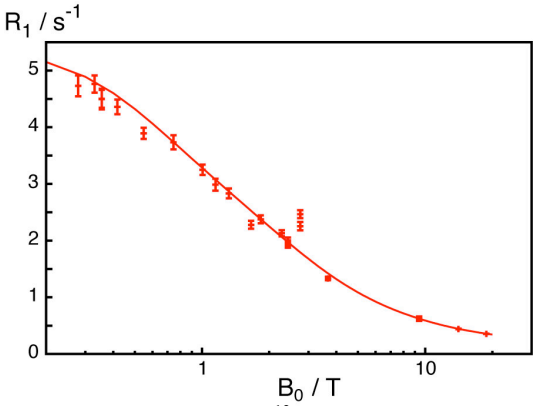

Figure 1. Longitudinal ^{13}C relaxation rates for the $^{13}C^1H^2H_2$ methyl δ1 group of residue Ile36 in ubiquitin. The solid line is calculated by fitting to local dynamics.

Finally, we show that high-resolution relaxometry offers a refined description of ps-ns motions in intrinsically disordered proteins (IDP's). Nitrogen-15 relaxation rates were measured between 0.22 and 22.3 T on the protein Artemis (Fig. 2). Relaxation rates at low field correlate with the degree of local ordering. The distribution of correlation times was reconstructed using regularization techniques.

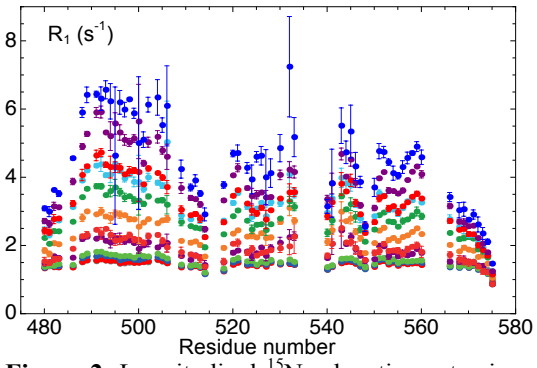

Figure 2. Longitudinal ^{15}N relaxation rates in the intrinsically disordered region of Artemis.

References

[1] A. G. Redfield, *Magn. Reson. Chem.*, 41, 753-768 (2003); [2] M. W. Clarkson, M. Lei, E. Z. Eisenmesser, W. Labeikovsky, A. Redfield and D. Kern, *J. Biomol. NMR*, 45, 217-225 (2009); [3] C. Charlier, S. N. Khan, T. Marquardsen, P. Pelupessy, V. Reiss, D. Sakellariou, G. Bodenhausen, F. Engelke and F. Ferrage, *J. Am. Chem. Soc.*, 135, 18665-18672 (2013).