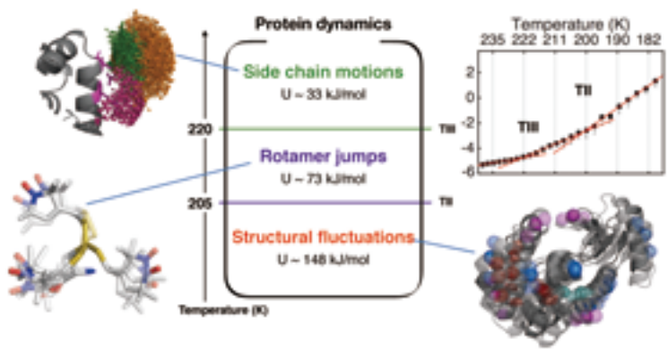
Professor Yun-Wei Chiang / ywchiang@gapp.nthu.edu.tw

Protein Has an Intrinsic Dynamic Component, Not Slaved by Solvent Motions

Protein structures are dynamic rather than static. They fluctuate on many different time scales and continually switch among conformational states to execute a variety of functions. Protein dynamics can span over a wide time range from shorter than nanoseconds to longer than milliseconds even at subfreezing temperatures. It is well-accepted that protein functions are governed by their dynamic motions. However, the role of solvent dynamics in the protein structure dynamics function relationship remains unclear. While solvent is essential for protein dynamics and function, its role in regulating the dynamics remains highly debated. This study aims to explore, in a fully hydrated condition, the connection between the protein dynamics and the dynamics of the surrounding solvent.

This study employs saturation transfer electron spin resonance (ST-ESR) to explore the issue and maps out the variation in protein local dynamics in the time range from microseconds to seconds, hence providing new insights into the dynamics on a longer time scale than has been extensively explored. We first demonstrate the reliability of ST-ESR by showing that the dynamical transition reported in the spectra coincides with liquid-liquid transition (LLT) in bulk solvents, accordingly establishing the connection between the dynamical changeover and the LLT of bulk solvent.

To investigate protein dynamics, site-directed spin labeling (SDSL) in combination with ST-ESR is introduced to explore the slow dynamics on the time range from microsecond to seconds in a fully hydrated protein over the temperature range 180 240 K. ST-ESR is developed to study the rotational dynamics on the very slow time scale (> μ s), where conventional ESR is not sensitive. The basic principle of ST-ESR is to collect the spectra under saturation conditions and high modulation amplitudes to observe the response of the spin system on spectral diffusion of saturation by molecular motions. SDSL in combination with ESR techniques is a powerful tool to explore local information on dynamics of molecular structure in an ensemble system. Basically, change in the dynamic structure of solvent can be probed by a nitroxide radical doped in the solvent. As opposed to other techniques detecting the overall dynamic motions in the ensemble, the SDSL-ESR provides local information on the dynamics of spin probes and the corresponding potential energy associated with the local environment in the solvent. When the spin-label side chain is incorporated into a protein by the SDSL methods, the tethered probe is also sensitive to various dynamic components related to the protein, such as dynamics of the side chain, backbone fluctuations, and interactions between tertiary structures. In this regard, the local environment reported on the ESR spectra of spin- labeled proteins is a composite of information from protein and nearby solvent molecules.



Spin-label ESR reveals the hierarchy of protein dynamics from μs to s. This study identifies the slow and collective dynamic component, intrinsic to protein and not slaved by solvent.

We show that the bulk solvent (10 mol % glycerol/ water) dynamics can only dominate the dynamics of the highly exposed sites in T4L below transition TI (∼ 190 K) in liquid II state. For other sites that are relatively less exposed, protein maintains control over the dynamics itself throughout the temperature range studied. The temperature-dependent behaviors of the protein-related dynamic components are not dominated by the solvent dynamics. The dynamics of spin labels is shown to reflect the overall structural dynamics in T4L (180 205 K), the dynamics of rotamer clusters (205 220 K), and the internal side-chain dynamics (220 240 K). The overall structural dynamics is collective and independent of protein structural segments, providing information for understanding the fundamental dynamic component of a protein that has not been reported. More than one protein-related dynamical transition is revealed. However, these dynamic components are arrested in the dehydration state. This study not only reveals the hierarchy of the protein dynamics associated with side-chain motions, but also provides quantitative descriptions for the dynamic components observed in the ST-ESR results of the fully hydrated T4L. The presence of hydration is required for protein to exhibit its dynamics, whereas it does not dominate the protein dynamics. The studies presented here support that bulk solvent plasticizes protein and facilitates rather than slaves protein dynamics.

(from left) Dr. Y.H. Kou, Dr. T.C. Sung, Professor Y.W. Chiang.

Research Highlights

- (2016) Ta-You Wu Memorial Award of Ministry of Science and Technology (MOST) of Taiwan
- (2013-2019) MOST Grant for Outstanding Early-career Researcher
- (2015) Outstanding Young Investigator Award of Taiwan Chemical Society

Research Output

- Y.H. Kuo, Y.W. Chiang*, Slow Dynamics around a Protein and Its Coupling to Solvent, ACS Central Science, 4 (2018) 645-655.
- T.Y. Kao, C.J. Tsai, Y.J. Lan, Y.W. Chiang*, Role of Conformational Heterogeneity in Regulating the Apoptotic Activity of BAX Protein, Physical Chemistry Chemical Physics, 19 (2017) 9584-9591.
- C.C. Wang, H.C. Chang, Y.C. Lai, H. Fang, C.C. Li, H.K. Hsu, Z.Y. Li, T.S. Lin, T.S. Kuo, F. Neese, S. Ye*, Y.W. Chiang*, M.L. Tsai*, W.F. Liaw*, W.Z. Lee*, A Structurally Characterized Nonheme Cobalt-Hydroperoxo Complex Derived from its Superoxo Intermediate via Hydrogen Atom Abstraction, Journal of the American Chemical Society, 138 (2016) 14186-14189.