Supporting Data

Aromatic Ring Flips Reveal Reshaping of Protein Dynamics in Crystals and Complexes

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IgG_GB1_complex_MODEL

PDB file containing the model of $GB1_{QDD}$ in complex with IgG that was used for Figure 1F-G. The model is based on Lamley *et al.*, 2014(*1*) and the PDB structures 1FCC, 1IGY, and 1IGC, as well as the new structure of $GB1_{QDD}$ (9I2I).

spectra

Spectra shown in the manuscript in .ucsf format. Detailed information on the acquisition parameters is provided in Table S2. Please note that there is a discrepancy in the referencing of carbon dimensions between these spectra files and the figures in the manuscript. The .ucsf spectra should be shifted by 2.66 ppm to have the correct referencing.

EXSY

Analysis of EXSY experiments at three different conditions. Each folder contains the following:

- **EXSY_Analysis.ipynb**: analysis script for peak fitting, intensity extraction, and fitting of exchange rate constants. It takes the following three scripts/folders as input.
- **EXSY_fit_new.py**: Equations used for the combined EXSY fit according to Farrow *et al.*, 1994 (*2*).
- ***_ucsf**: spectra as 2D planes (alternating exchange and reference experiment with the same exchange time) in *.ucsf format and the **vdlist** that defines the exchange time (see supporting information for details).
- **R1_225K_700MHz_39kHz_output.csv**: R_1 relaxation rate constants used for the fit.
- **output**: output of the analysis script
- the remaining files in these folders are outputs that were used to generate the figures shown in the manuscript.

In **EXSY_225K_700MHz_39kHz**, two folders named *_ucsf exist. This experiment was measured twice, summed up before the analysis with the script **add_spectra.ipynb**, and stored in **sum_ucsf**.

order_parameters

Analysis of order parameters for **crystal** and **complex**.
The folder structure of **REDOR_*** is as follows:

- **REDOR_Analysis.ipynb**: analysis script for peak fitting, intensity extraction, and fitting of REDOR curves. It takes the following three folders as input.
- **300_ucsf**: spectra of the recoupling experiment as 2D planes in *.ucsf format and the **vdlist** that defines the number of rotor periods (see supporting information for details).
- **301_ucsf**: spectra of the reference experiment as 2D planes in *.ucsf format and the **vdlist** that defines the number of rotor periods (see supporting information for details)
- **GAMMA_data**: Simulated REDOR curves.
- **output**: output of the analysis script

The script **plotting_fits.ipynb** plots the data from the analysis as shown in the manuscript and saves the files into **plots**.

relaxation_rates

Analysis of R_1 and $R_{1\rho}$ relaxation rate constants for the backbone, the complex, and the crystal at two different temperatures. Within these folders, each experiment is analyzed inside its own folder, starting with either **R1_*** or **R1rho_***. Additionally, **plotting_fits.ipynb** is a script to collect data from the experiment folders, plot the fits and rates as shown in Figures S11, S12, and S16-S18 (saved in **plots**), and create input scripts for the detectors analysis (saved in **R1rho_data**). The experiment folders are structured as follows:

- **R1(p)_Analysis_1D.ipynb**: analysis script for peak fitting, intensity extraction, and fitting of rate constants. It takes the following two scripts/folders as input.
- **spectra_handling.py**: custom functions for handling of spectra based on the nmrglue package(*3*).
- ***_ucsf**: spectra as 2D planes in *.ucsf format and the **vdlist** that defines the relaxation time (see supporting information for details).
- **output**: output of the analysis script

Note that, in the analysis of the aromatics in the crystal (**aromatics_crystal_288K** and **aromatics_crystal_304K**), the labels of the residues are not correct due to a mistake in an earlier version of the assignment (Y3 and Y33 are switched). This is corrected in the **plotting_fits.ipynb** script for the following detectors analysis.
For the fit of the backbone spectra, peaks were picked and fitted based on the assignment instead of using the built-in nmrglue function due to the complexity of the backbone spectrum. The resulting nmrglue peak list was exported and used for the analysis of the relaxation experiments. The script for this can be found in **backbone/peaks/pick_Peaks.ipynb**

detectors

Input and output of the detectors analysis of **backbone** and **aromatics** relaxation data. Data was analyzed with the Detectorist package available at https://github.com/ThatPerson/Detectorist (*4*).
.toml** files are input scripts that define parameters for the analysis, the location of the files containing the experimental relaxation rate constants, and the folders for the output. The files with relaxation rate constants are in the folders **13C**, **13C_IgG**, and **15N**. Output files are in **fits_.

thermodynamic_parameters

experimental contains the experimental exchange rate constants for Y3 and Y45 (datapoints.txt) and the results of the combined Eyring fit (parameters.txt).
free_energy_profiles_MD contains the raw data for the free energy profiles shown in Figure 5D for the three tyrosines for solution and crystal, respectively.

distance_traces_MD

The folders **solution** and **crystal** contain all traces of distances and angles from MD simulations that are shown in Figures 6C, S20, and S21. The file format (.npy) is a 1D numpy array with either the distance in Å or the angle in ° for each frame of the simulation.

References

(*1*) Lamley, J. M. et al. (2014). Solid-state NMR of a protein in a precipitated complex with a full-length antibody. *Journal of the American Chemical Society 136*, 16800–16806.

(*2*) Farrow, N. A. et al. (1994). A heteronuclear correlation experiment for simultaneous determination of 15N longitudinal decay and chemical exchange rates of systems in slow equilibrium. *Journal of Biomolecular NMR 4*, 727–734.

(*3*) Helmus, J. J., and Jaroniec, C. P. (2013). Nmrglue: An open source Python package for the analysis of multidimensional NMR data. *Journal of Biomolecular NMR 55*, 355–367.

(*4*) Tatman, B. P. The development of methods to study structure and dynamics in biological systems using solid-state NMR, Available at `https://wrap.warwick.ac.uk/id/eprint/185489/`, PhD thesis, University of Warwick, 2023.