Supporting Information for

Regulating conformation of prion protein through ligand binding

Norifumi Yamamoto[a,b] and Kazuo Kuwata[a,b,*]

[a]*Division of Prion Research, Center for Emerging Infectious Diseases,*

Gifu University, 1-1 Yanagido, Gifu 501-1194, Japan

[b]*CREST Project, Japan Science and Technology Agency, 4-1-8 Honcho,*

Kawaguchi, Saitama 332-0012, Japan

[*]Corresponding author: Tel: +81-58-230-6145; Fax: +81-58-230-6144; E-mail: kuwata@gifu-u.ac.jp

Simulation details

All MD simulations were performed using the Gromacs 4.0.4 package[1] with the Gromos96 43a2 force field.[2] The particle-mesh Ewald scheme was used to treat the nonbonded electrostatic interactions, where a Fourier grid spacing of 0.135 nm was used. The cut-off scheme was used to treat the nonbonded vdW interactions. The long-range cut-off lengths were set as 1.0 and 1.4 nm for the electrostatic and vdW interactions, respectively. The list of nonbonded interactions was truncated at 1 nm. The modeled systems were energy minimized using the steepest descent method with no constraints. Starting with the energy-minimized structures, position-restrained MD simulations were carried out for a period of 200 ps using the linear constraint solver (LINCS) algorithm, where lengths of all bonds in the systems were constrained. After position-restrained simulations, MD simulations with no constraints were conducted. All simulations were performed in the *NpT* ensemble, where constant temperature was maintained at 300 K using the Berendsen-type temperature-coupling scheme with a time constant of τ_T = 0.1 ps, and constant pressure was maintained at 1 atm using the Berendsen-type pressure-coupling scheme with a time constant of τ_p = 0.5 ps. The isotropic compressibility was set to a value of 4.5×10^{-5} bar^{-1}. Initial velocities were obtained randomly from a Maxwellian distribution at 300 K. An integration time step of 2 fs was used for all MD simulations. Protein and its complex were placed in a truncated octahedron box, where the minimal distance between protein and box walls was set as 1.0 nm, and periodic boundary conditions were used. The SPC/E water model[3] and Smith's urea model[4] were employed to describe the solvents. Appropriate counterions (Na^+ or Cl^-) were added. The urea-driven denaturing simulation of the monomeric PrP was performed in an aqueous urea solution with 5229 water molecules and 935 urea molecules,

corresponding to a urea mole fraction of 0.15 and a concentration of 5.88 M. The urea-driven denaturing simulation of PrP−GN8 was performed in an aqueous urea solution with 5498 water molecules and 990 urea molecules, corresponding to a urea mole fraction of 0.15 and a concentration of 5.91 M. During these simulations, configurations were saved every 1 ps.

GN8, *N*,*N*'-(methylenedi-4,1-phenylene)bis[2-(1-pyrrolidinyl)acetamide],[5] is a typical alkaloid with a pyrrolidine ring, which is protonated and positively charged at physiological pH. Thus, in our calculation, a doubly protonated cationic form corresponding to a chemical structure of $(C_{25}H_{34}N_4O_2)^{2+}$ was used. We employed the Gromos96 topology data generated by the Dundee PRODRG 2.5 Server.[6] Partial charges were determined by fitting the electrostatic potential derived from a quantum chemistry calculation performed at the DFT-B3LYP/cc-pVDZ level using the Gaussian 03 (revision D.01) program,[7] according to the Merz–Singh–Kollman scheme.[8,9]

(1) Hess, B.; Kutzner, C.; van der Spoel, D.; Lindahl, E. *J. Chem. Theory Comput.* **2008**, *4*, 435.

(2) Schuler, L. D.; van Gunsteren, W. F. *Mol. Simul.* **2000**, *25*, 301.

(3) Berendsen, H. J. C.; Grigera, J. R.; Straatsma, T. P. *J. Phys. Chem.* **1987**, *91*, 6269.

(4) Smith, L. J.; Berendsen, H. J. C.; van Gunsteren, W. F. *J. Phys. Chem. B* **2004**, *108*, 1065.

(5) Kuwata, K.; Nishida, N.; Matsumoto, T.; Kamatari, Y. O.; Hosokawa-Muto, J.; Kodama, K.; Nakamura, H. K.; Kimura, K.; Kawasaki, M.; Takakura, Y.; Shirabe, S.; Takata, J.; Kataoka, Y.; Katamine, S. *Proc. Natl. Acad. Sci. USA* **2007**, *104*, 11921.

(6) Schüttelkopf, A. W.; van Aalten, D. M. F. *Acta Cryst.* **2004**, *D60*, 1355.

(7) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Montgomery, J. A., Jr.; Vreven, T.; Kudin, K. N.; Burant, J. C.; Millam, J. M.; Iyengar, S. S.; Tomasi, J.; Barone, V.; Mennucci, B.; Cossi, M.; Scalmani, G.; Rega, N.; Petersson, G. A.; Nakatsuji, H.; Hada, M.; Ehara, M.; Toyota, K.; Fukuda, R.; Hasegawa, J.; Ishida, M.; Nakajima, T.; Honda, Y.; Kitao, O.; Nakai, H.; Klene, M.; Li, X.; Knox, J. E.; Hratchian, H. P.; Cross, J. B.; Bakken, V.; Adamo, C.; Jaramillo, J.; Gomperts, R.; Stratmann, R. E.; Yazyev, O.; Austin, A. J.; Cammi, R.; Pomelli, C.; Ochterski, J. W.; Ayala, P. Y.; Morokuma, K.; Voth, G. A.; Salvador, P.; Dannenberg, J. J.; Zakrzewski, V. G.; Dapprich, S.; Daniels, A. D.; Strain, M. C.; Farkas, O.; Malick, D. K.; Rabuck, A. D.; Raghavachari, K.; Foresman, J. B.; Ortiz, J. V.; Cui, Q.; Baboul, A. G.; Clifford, S.; Cioslowski, J.; Stefanov, B. B.; Liu, G.; Liashenko, A.; Piskorz, P.; Komaromi, I.; Martin, R. L.; Fox, D. J.; Keith, T.; Al-Laham, M. A.; Peng, C. Y.; Nanayakkara, A.; Challacombe, M.; Gill, P. M. W.; Johnson, B.; Chen, W.; Wong, M. W.; Gonzalez, C.; Pople, J. A. Gaussian 03, Revision D.02; Gaussian, Inc.: Wallingford CT, 2004.

(8) Singh, U. C.; Kollman, P. A. *J. Comput. Chem.* **1984**, *5*, 129.

(9) Besler, B. H.; Merz, K. M., Jr.; Kollman, P. A. *J. Comput. Chem.* **1990**, *11*, 431.