



Universiteit
Leiden
The Netherlands

Classical and paramagnetic NMR spectroscopy techniques applied to different protein systems

Skinner, S.P.

Citation

Skinner, S. P. (2013, December 5). *Classical and paramagnetic NMR spectroscopy techniques applied to different protein systems*. Retrieved from <https://hdl.handle.net/1887/22714>

Version: Not Applicable (or Unknown)

License: [Licence agreement concerning inclusion of doctoral thesis in the Institutional Repository of the University of Leiden](#)

Downloaded from: <https://hdl.handle.net/1887/22714>

Note: To cite this publication please use the final published version (if applicable).

Cover Page



Universiteit Leiden



The handle <http://hdl.handle.net/1887/22714> holds various files of this Leiden University dissertation.

Author: Skinner, Simon Peter

Title: Classical and paramagnetic NMR spectroscopy techniques applied to different protein systems

Issue Date: 2013-12-05

Reference List

1. Kundu, S., Melton, J. S., Sorensen, D. C., and Phillips, G. N., Jr. (2002) Dynamics of proteins in crystals: comparison of experiment with simple models, *Biophys J* 83, 723-732.
2. Wuthrich, K. (1986) *NMR of Proteins and Nucleic Acids*, 1 ed., Wiley-Interscience, New York.
3. Driscoll, P. C., Clore, G. M., Marion, D., Wingfield, P. T., and Gronenborn, A. M. (1990) Complete resonance assignment for the polypeptide backbone of interleukin 1 beta using three-dimensional heteronuclear NMR spectroscopy, *Biochemistry* 29, 3542-3556.
4. Driscoll, P. C., Gronenborn, A. M., Wingfield, P. T., and Clore, G. M. (1990) Determination of the secondary structure and molecular topology of interleukin-1 beta by use of two- and three-dimensional heteronuclear ¹⁵N-¹H NMR spectroscopy, *Biochemistry* 29, 4668-4682.
5. Marion, D., Driscoll, P. C., Kay, L. E., Wingfield, P. T., Bax, A., Gronenborn, A. M., and Clore, G. M. (1989) Overcoming the overlap problem in the assignment of ¹H NMR spectra of larger proteins by use of three-dimensional heteronuclear ¹H-¹⁵N Hartmann-Hahn-multiple quantum coherence and nuclear Overhauser-multiple quantum coherence spectroscopy: application to interleukin 1 beta, *Biochemistry* 28, 6150-6156.
6. Wijmenga, S. S., Hallenga, K., and Hilbers, C. W. (1989) A three-dimensional heteronuclear multiple-quantum coherence homonuclear hartmann-hahn experiment, *J Mag Reson* 84, 634-642.
7. Fesik, S. W., and Zuiderweg, E. R. P. (1988) Heteronuclear three-dimensional nmr spectroscopy. A strategy for the simplification of homonuclear two-dimensional NMR spectra, *J Magn Reson* 78, 588-593.
8. Shon, K., and Opella, S. J. (1989) Detection of ¹H homonuclear NOE between amide sites in proteins with ¹H-¹⁵N heteronuclear correlation spectroscopy, *J Magn Reson* 82, 193-197.
9. Grzesiek, S., and Bax, A. (1992) An efficient experiment for sequential backbone assignment of medium-sized isotopically enriched proteins, *J Magn Reson* 99, 201-207.
10. Clubb, R. T., Thanabal, V., and Wagner, G. (1992) A constant-time three-dimensional triple-resonance pulse scheme to correlate intraresidue ¹HN, ¹⁵N, and ¹³C' chemical shifts in ¹⁵N-¹³C-labelled proteins, *J Magn Reson* 97, 213-217.
11. Sattler, M., Schleucher, J. r., and Griesinger, C. (1999) Heteronuclear multidimensional NMR experiments for the structure determination of proteins in solution employing pulsed field gradients, *Prog Nucl Magn Reson Spectrosc* 34, 93-158.
12. Yamazaki, T., Lee, W., Arrowsmith, C. H., Muhandiram, D. R., and Kay, L. E. (1994) A Suite of Triple Resonance NMR Experiments for the Backbone Assignment of ¹⁵N, ¹³C, ²H Labeled Proteins with High Sensitivity, *J Am Chem Soc* 116, 11655-11666.
13. Kay, L. E., Ikura, M., Tschudin, R., and Bax, A. (1990) Three-dimensional triple-resonance NMR spectroscopy of isotopically enriched proteins, *J Magn Reson* 89, 496-514.
14. Pervushin, K., Riek, R., Wider, G., and Wuthrich, K. (1997) Attenuated T2 relaxation by mutual cancellation of dipole-dipole coupling and chemical shift anisotropy indicates an avenue to NMR structures of very large biological macromolecules in solution, *Proc Natl Acad Sci U S A* 94, 12366-12371.
15. Keeler, J. (2010) *Understanding NMR spectroscopy*, 2nd ed., Wiley-Blackwell.
16. Fernandez, C., and Wider, G. (2003) TROSY in NMR studies of the structure and function of large biological macromolecules, *Curr Opin Struct Biol* 13, 570-580.

17. Lescop, E., Schanda, P., and Brutscher, B. (2007) A set of BEST triple-resonance experiments for time-optimized protein resonance assignment, *J Magn Reson* 187, 163-169.
18. Schanda, P., Van Melckebeke, H., and Brutscher, B. (2006) Speeding up three-dimensional protein NMR experiments to a few minutes, *J Am Chem Soc* 128, 9042-9043.
19. Pervushin, K., Vogeli, B., and Eletsky, A. (2002) Longitudinal (1)H relaxation optimization in TROSY NMR spectroscopy, *J Am Chem Soc* 124, 12898-12902.
20. Schanda, P., Kupce, E., and Brutscher, B. (2005) SOFAST-HMQC experiments for recording two-dimensional heteronuclear correlation spectra of proteins within a few seconds, *J Biomol NMR* 33, 199-211.
21. Schanda, P., and Brutscher, B. (2006) Hadamard frequency-encoded SOFAST-HMQC for ultrafast two-dimensional protein NMR, *J Magn Reson* 178, 334-339.
22. Schanda, P., and Brutscher, B. (2005) Very fast two-dimensional NMR spectroscopy for real-time investigation of dynamic events in proteins on the time scale of seconds, *J Am Chem Soc* 127, 8014-8015.
23. Tugarinov, V., Choy, W. Y., Orekhov, V. Y., and Kay, L. E. (2005) Solution NMR-derived global fold of a monomeric 82-kDa enzyme, *Proc Natl Acad Sci U S A* 102, 622-627.
24. Guntert, P. (2004) Automated NMR structure calculation with CYANA, *Methods Mol Biol* 278, 353-378.
25. Herrmann, T., Guntert, P., and Wuthrich, K. (2002) Protein NMR structure determination with automated NOE assignment using the new software CANDID and the torsion angle dynamics algorithm DYANA, *J Mol Biol* 319, 209-227.
26. Bertini, I., Luchinat, C., and Parigi, G. (2002) Magnetic susceptibility in paramagnetic NMR, *Prog Nucl Magn Reson Spectrosc* 40, 249-273.
27. Keizers, P. H., Saragliadis, A., Hiruma, Y., Overhand, M., and Ubbink, M. (2008) Design, synthesis, and evaluation of a lanthanide chelating protein probe: CLaNP-5 yields predictable paramagnetic effects independent of environment, *J Am Chem Soc* 130, 14802-14812.
28. Schmitz, C., Stanton-Cook, M. J., Su, X. C., Otting, G., and Huber, T. (2008) Numbat: an interactive software tool for fitting Deltachi-tensors to molecular coordinates using pseudocontact shifts, *J Biomol NMR* 41, 179-189.
29. Solomon, I. (1955) Relaxation Processes in a System of Two Spins, *Phys Rev* 99, 559-565.
30. Bloembergen, N., and Morgan, L. O. (1961) Proton Relaxation Times in Paramagnetic Solutions. Effects of Electron Spin Relaxation, *J Chem Phys* 34, 842-850.
31. Volkov, A. N., Worrall, J. A., Holtzmann, E., and Ubbink, M. (2006) Solution structure and dynamics of the complex between cytochrome c and cytochrome c peroxidase determined by paramagnetic NMR, *Proc Natl Acad Sci U S A* 103, 18945-18950.
32. Scanu, S., Forster, J., Finiguerra, M. G., Shabestari, M. H., Huber, M., and Ubbink, M. (2012) The complex of cytochrome f and plastocyanin from Nostoc sp. PCC 7119 is highly dynamic, *Chembiochem* 13, 1312-1318.
33. Iwahara, J., Schwieters, C. D., and Clore, G. M. (2004) Ensemble approach for NMR structure refinement against (1)H paramagnetic relaxation enhancement data arising from a flexible paramagnetic group attached to a macromolecule, *J Am Chem Soc* 126, 5879-5896.
34. Gueron, M. (1975) Nuclear relaxation in macromolecules by paramagnetic ions: a novel mechanism, *J Magn Reson* 19, 58-66.
35. Kowalsky, A. (1965) Nuclear Magnetic Resonance Studies of Cytochrome c. Possible Electron Delocalization*, *Biochemistry* 4, 2382-2388.
36. Wuthrich, K. (1969) High-resolution proton nuclear magnetic resonance spectroscopy of cytochrome, *Proc Natl Acad Sci U S A* 63, 1071-1078.

37. Moura, J. J. G., Xavier, A. n. V., Bruschi, M., and Gall, J. L. (1977) NMR characterization of three forms of ferredoxin from *Desulphovibrio gigas*, a sulphate reducer, *Biochim Biophys Acta* 459, 278-289.
38. Salmeen, I., and Palmer, G. (1972) Contact-shifted NMR of spinach ferredoxin: Additional resonances and partial assignments, *Arch Biochem Biophys* 150, 767-773.
39. Oh, B. H., Westler, W. M., Darba, P., and Markley, J. L. (1988) Protein carbon-13 spin systems by a single two-dimensional nuclear magnetic resonance experiment, *Science* 240, 908-911.
40. Phillips, W. D., Poe, M., Weiher, J. F., McDonald, C. C., and Lovenberg, W. (1970) Proton magnetic resonance, magnetic susceptibility and Mossbauer studies of *Clostridium pasteurianum* rubredoxin, *Nature* 227, 574-577.
41. Ubbink, M., Ejdeback, M., Karlsson, B. G., and Bendall, D. S. (1998) The structure of the complex of plastocyanin and cytochrome f, determined by paramagnetic NMR and restrained rigid-body molecular dynamics, *Structure* 6, 323-335.
42. Salgado, J., Jimenez, H. R., Moratal, J. M., Kroes, S., Warmerdam, G. C. M., and Canters, G. W. (1996) Paramagnetic Cobalt and Nickel Derivatives of *Alcaligenes denitrificans* Azurin and Its M121Q Mutant. A ¹H NMR Study., *Biochemistry* 35, 1810-1819.
43. Piccioli, M., Luchinat, C., Mizoguchi, T. J., Ramirez, B. E., Gray, H. B., and Richards, J. H. (1995) Paramagnetic NMR spectroscopy and coordination structure of cobalt(II) Cys112Asp azurin, *Inorg Chem* 34, 737-742.
44. John, M., Pintacuda, G., Park, A. Y., Dixon, N. E., and Otting, G. (2006) Structure determination of protein-ligand complexes by transferred paramagnetic shifts, *J Am Chem Soc* 128, 12910-12916.
45. Feeney, J., Birdsall, B., Bradbury, A. F., Biekofsky, R. R., and Bayley, P. M. (2001) Calmodulin tagging provides a general method of using lanthanide induced magnetic field orientation to observe residual dipolar couplings in proteins in solution, *J Biomol NMR* 21, 41-48.
46. Ma, C., and Opella, S. J. (2000) Lanthanide ions bind specifically to an added "EF-hand" and orient a membrane protein in micelles for solution NMR spectroscopy, *J Magn Reson* 146, 381-384.
47. Johnson, P. E., Brun, E., MacKenzie, L. F., Withers, S. G., and McIntosh, L. P. (1999) The cellulose-binding domains from *Cellulomonas fimi* beta-1, 4-glucanase CenC bind nitroxide spin-labeled celooligosaccharides in multiple orientations, *J Mol Biol* 287, 609-625.
48. Saio, T., Ogura, K., Yokochi, M., Kobashigawa, Y., and Inagaki, F. (2009) Two-point anchoring of a lanthanide-binding peptide to a target protein enhances the paramagnetic anisotropic effect, *J Biomol NMR* 44, 157-166.
49. Su, X. C., McAndrew, K., Huber, T., and Otting, G. (2008) Lanthanide-binding peptides for NMR measurements of residual dipolar couplings and paramagnetic effects from multiple angles, *J Am Chem Soc* 130, 1681-1687.
50. Gaponenko, V., Dvoretzky, A., Walsby, C., Hoffman, B. M., and Rosevear, P. R. (2000) Calculation of z-coordinates and orientational restraints using a metal binding tag, *Biochemistry* 39, 15217-15224.
51. Barthelmes, K., Reynolds, A. M., Peisach, E., Jonker, H. R. A., DeNunzio, N. J., Allen, K. N., Imperiali, B., and Schwalbe, H. (2011) Engineering Encodable Lanthanide-Binding Tags into Loop Regions of Proteins, *J Am Chem Soc* 133, 808-819.
52. Pintacuda, G., Moshref, A., Leonchiks, A., Sharipo, A., and Otting, G. (2004) Site-specific labelling with a metal chelator for protein-structure refinement, *J Biomol NMR* 29, 351-361.
53. Dvoretzky, A., Gaponenko, V., and Rosevear, P. R. (2002) Derivation of structural restraints using a thiol-reactive chelator, *FEBS Lett* 528, 189-192.

54. Kamen, D. E., Cahill, S. M., and Girvin, M. E. (2007) Multiple alignment of membrane proteins for measuring residual dipolar couplings using lanthanide ions bound to a small metal chelator, *J Am Chem Soc* 129, 1846-1847.
55. Leonov, A., Voigt, B., Rodriguez-Castaneda, F., Sakhaei, P., and Griesinger, C. (2005) Convenient synthesis of multifunctional EDTA-based chiral metal chelates substituted with an S-mesylcysteine, *Chemistry* 11, 3342-3348.
56. Rodriguez-Castaneda, F., Haberz, P., Leonov, A., and Griesinger, C. (2006) Paramagnetic tagging of diamagnetic proteins for solution NMR, *Magn Reson Chem* 44 Spec No, S10-16.
57. Haussinger, D., Huang, J. R., and Grzesiek, S. (2009) DOTA-M8: An extremely rigid, high-affinity lanthanide chelating tag for PCS NMR spectroscopy, *J Am Chem Soc* 131, 14761-14767.
58. Su, X. C., Man, B., Beeren, S., Liang, H., Simonsen, S., Schmitz, C., Huber, T., Messerle, B. A., and Otting, G. (2008) A dipicolinic acid tag for rigid lanthanide tagging of proteins and paramagnetic NMR spectroscopy, *J Am Chem Soc* 130, 10486-10487.
59. Ikegami, T., Verdier, L., Sakhaei, P., Grimme, S., Pescatore, B., Saxena, K., Fiebig, K. M., and Griesinger, C. (2004) Novel techniques for weak alignment of proteins in solution using chemical tags coordinating lanthanide ions, *J Biomol NMR* 29, 339-349.
60. Haberz, P., Rodriguez-Castaneda, F., Junker, J., Becker, S., Leonov, A., and Griesinger, C. (2006) Two new chiral EDTA-based metal chelates for weak alignment of proteins in solution, *Org Lett* 8, 1275-1278.
61. Liu, W. M., Keizers, P. H., Hass, M. A., Blok, A., Timmer, M., Sarris, A. J., Overhand, M., and Ubbink, M. (2012) A pH-Sensitive, Colorful, Lanthanide-Chelating Paramagnetic NMR Probe, *J Am Chem Soc* 134, 17306-17313.
62. Banci, L., Bertini, I., Bren, K. L., Cremonini, M. A., Gray, H. B., Luchinat, C., and Turano, P. (1996) The use of pseudocontact shifts to refine solution structures of paramagnetic metalloproteins: Met80Ala cyano-cytochrome c as an example, *J. Biol. Inorg. Chem.* 1, 117-126.
63. Banci, L., Bertini, I., Savellini, G. G., Romagnoli, A., Turano, P., Cremonini, M. A., Luchinat, C., and Gray, H. B. (1997) Pseudocontact shifts as constraints for energy minimization and molecular dynamics calculations on solution structures of paramagnetic metalloproteins, *Proteins* 29, 68-76.
64. Schwieters, C. D., Kuszewski, J. J., Tjandra, N., and Clore, G. M. (2003) The Xplor-NIH NMR molecular structure determination package, *J Magn Reson* 160, 65-73.
65. Banci, L., Bertini, I., Cavallaro, G., Giachetti, A., Luchinat, C., and Parigi, G. (2004) Paramagnetism-based restraints for Xplor-NIH, *J Biomol NMR* 28, 249-261.
66. Pintacuda, G., Keniry, M. A., Huber, T., Park, A. Y., Dixon, N. E., and Otting, G. (2004) Fast structure-based assignment of ¹⁵N HSQC spectra of selectively ¹⁵N-labeled paramagnetic proteins, *J Am Chem Soc* 126, 2963-2970.
67. Schmitz, C., John, M., Park, A. Y., Dixon, N. E., Otting, G., Pintacuda, G., and Huber, T. (2006) Efficient chi-tensor determination and NH assignment of paramagnetic proteins, *J Biomol NMR* 35, 79-87.
68. John, M., Schmitz, C., Park, A. Y., Dixon, N. E., Huber, T., and Otting, G. (2007) Sequence-specific and stereospecific assignment of methyl groups using paramagnetic lanthanides, *J Am Chem Soc* 129, 13749-13757.
69. Allerhand, A., Doddrell, D., Glushko, V., Cochran, D. W., Wenkert, E., Lawson, P. J., and Gurd, F. R. (1971) Conformation and segmental motion of native and denatured ribonuclease A in solution. Application of natural-abundance carbon-13 partially relaxed Fourier transform nuclear magnetic resonance, *J Am Chem Soc* 93, 544-546.
70. Nigen, A. M., Keim, P., Marshall, R. C., Morrow, J. S., and Gurd, F. R. (1972) Carbon 13 nuclear magnetic resonance spectroscopy of myoglobins and ribonuclease A carboxymethylated with enriched (2- ¹³C)bromoacetate, *J Biol Chem* 247, 4100-4102.

71. Glushko, V., Lawson, P. J., and Gurd, F. R. (1972) Conformational states of bovine pancreatic ribonuclease A observed by normal and partially relaxed carbon 13 nuclear magnetic resonance, *J Biol Chem* 247, 3176-3185.
72. Jones, W. C., Rothgeb, T. M., and Gurd, F. R. (1976) Nuclear magnetic resonance studies of sperm whale myoglobin specifically enriched with ¹³C in the methionine methyl groups, *J Biol Chem* 251, 7452-7460.
73. Campbell, I. D., Dobson, C. M., and Williams, R. J. (1985) The study of conformational states of proteins by nuclear magnetic resonance, *Biochem J* 231, 1-10.
74. Hughes, L. T., Cohen, J. S., Szabo, A., Niu, C., and Matsuura, S. (1984) ¹³C NMR studies of the molecular dynamics of selectively ¹³C-enriched ribonuclease complexes, *Biochemistry* 23, 4390-4394.
75. Rozovsky, S., Jogl, G., Tong, L., and McDermott, A. E. (2001) Solution-state NMR investigations of triosephosphate isomerase active site loop motion: ligand release in relation to active site loop dynamics, *J Mol Biol* 310, 271-280.
76. Seifert, M. H., Breitenlechner, C. B., Bossemeyer, D., Huber, R., Holak, T. A., and Engh, R. A. (2002) Phosphorylation and flexibility of cyclic-AMP-dependent protein kinase (PKA) using (³¹P) NMR spectroscopy, *Biochemistry* 41, 5968-5977.
77. Kay, L. E., Torchia, D. A., and Bax, A. (1989) Backbone dynamics of proteins as studied by ¹⁵N inverse detected heteronuclear NMR spectroscopy: application to staphylococcal nuclease, *Biochemistry* 28, 8972-8979.
78. Lipari, G., and Szabo, A. (1982) Model-free approach to the interpretation of nuclear magnetic resonance relaxation in macromolecules. 1. Theory and range of validity, *J Am Chem Soc* 104, 4546-4559.
79. Lipari, G., and Szabo, A. (1982) Model-free approach to the interpretation of nuclear magnetic resonance relaxation in macromolecules. 2. Analysis of experimental results, *J Am Chem Soc* 104, 4559-4570.
80. Dayie, K. T., and Wagner, G. (1997) Carbonyl Carbon Probe of Local Mobility in ¹³C,¹⁵N-Enriched Proteins Using High-Resolution Nuclear Magnetic Resonance, *J Am Chem Soc* 119, 7797-7806.
81. Yamazaki, T., Muhandiram, R., and Kay, L. E. (1994) NMR Experiments for the Measurement of Carbon Relaxation Properties in Highly Enriched, Uniformly ¹³C,¹⁵N-Labeled Proteins: Application to ¹³C alpha carbons, *J Am Chem Soc* 116, 8266-8278.
82. Wang, T., Cai, S., and Zuiderweg, E. R. (2003) Temperature dependence of anisotropic protein backbone dynamics, *J Am Chem Soc* 125, 8639-8643.
83. Fischer, M. W., Zeng, L., Majumdar, A., and Zuiderweg, E. R. (1998) Characterizing semilocal motions in proteins by NMR relaxation studies, *Proc Natl Acad Sci U S A* 95, 8016-8019.
84. Pang, Y., Buck, M., and Zuiderweg, E. R. (2002) Backbone dynamics of the ribonuclease binase active site area using multinuclear (¹⁵N and (¹³CO) NMR relaxation and computational molecular dynamics, *Biochemistry* 41, 2655-2666.
85. Desvaux, H., and Berthault, P. (1999) Study of dynamic processes in liquids using off-resonance rf irradiation, *Prog Nucl Magn Reson Spectrosc* 35, 295-340.
86. Palmer, A. G., 3rd, Kroenke, C. D., and Loria, J. P. (2001) Nuclear magnetic resonance methods for quantifying microsecond-to-millisecond motions in biological macromolecules, *Methods Enzymol* 339, 204-238.
87. Deverell, C., Morgan, R. E., and Strange, J. H. (1970) Studies of chemical exchange by nuclear magnetic relaxation in the rotating frame, *Mol Phys* 18, 553-559.
88. Meiboom, S. (1961) Nuclear Magnetic Resonance Study of the Proton Transfer in Water, *J Chem Phys* 34, 375-388.
89. Szyperski, T., Luginbuhl, P., Otting, G., Guntert, P., and Wuthrich, K. (1993) Protein dynamics studied by rotating frame ¹⁵N spin relaxation times, *J Biomol NMR* 3, 151-164.

90. Akke, M., and Palmer, A. G. (1996) Monitoring Macromolecular Motions on Microsecond to Millisecond Time Scales by R1 ρ -R1 Constant Relaxation Time NMR Spectroscopy, *J Am Chem Soc* 118, 911-912.
91. Auer, R., Neudecker, P., Muhandiram, D. R., Lundstrom, P., Hansen, D. F., Konrat, R., and Kay, L. E. (2009) Measuring the Signs of ¹H α Chemical Shift Differences Between Ground and Excited Protein States by Off-Resonance Spin-Lock R1 ρ NMR Spectroscopy, *J Am Chem Soc* 131, 10832-10833.
92. Hansen, A. L., Nikolova, E. N., Casiano-Negroni, A., and Al-Hashimi, H. M. (2009) Extending the Range of Microsecond-to-Millisecond Chemical Exchange Detected in Labeled and Unlabeled Nucleic Acids by Selective Carbon R1 ρ NMR Spectroscopy, *J Am Chem Soc* 131, 3818-3819.
93. Korzhnev, D., Orekhov, V., Dahlquist, F., and Kay, L. (2003) Off-resonance R1 ρ relaxation outside of the fast exchange limit: An experimental study of a cavity mutant of T4 lysozyme, *J Biomol NMR* 26, 39-48.
94. Korzhnev, D. M., Orekhov, V. Y., and Kay, L. E. (2005) Off-Resonance R1 ρ NMR Studies of Exchange Dynamics in Proteins with Low Spin-Lock Fields. An Application to a Fyn SH3 Domain, *J Am Chem Soc* 127, 713-721.
95. Lundström, P., and Akke, M. (2005) Off-resonance rotating-frame amide proton spin relaxation experiments measuring microsecond chemical exchange in proteins, *J Biomol NMR* 32, 163-173.
96. Massi, F., Grey, M. J., and Palmer, A. G., 3rd. (2005) Microsecond timescale backbone conformational dynamics in ubiquitin studied with NMR R1 ρ relaxation experiments, *Protein Sci* 14, 735-742.
97. Massi, F., Johnson, E., Wang, C., Rance, M., and Palmer, A. G., 3rd. (2004) NMR R1 ρ rotating-frame relaxation with weak radio frequency fields, *J Am Chem Soc* 126, 2247-2256.
98. Baldwin, A. J., and Kay, L. E. (2013) An R1 ρ expression for a spin in chemical exchange between two sites with unequal transverse relaxation rates, *J Biomol NMR* 55, 211-218.
99. Carr, H. Y., and Purcell, E. M. (1954) Effects of Diffusion on Free Precession in Nuclear Magnetic Resonance Experiments, *Phys Rev* 94, 630-638.
100. Luz, Z., and Meiboom, S. (1963) Nuclear Magnetic Resonance Study of the Protolysis of Trimethylammonium Ion in Aqueous Solution---Order of the Reaction with Respect to Solvent, *J Chem Phys* 39, 366-370.
101. Meiboom, S., and Gill, D. (1958) Modified Spin-Echo Method for Measuring Nuclear Relaxation Times, *Rev Sci Instrum* 29, 688-691.
102. Mulder, F. A., Skrynnikov, N. R., Hon, B., Dahlquist, F. W., and Kay, L. E. (2001) Measurement of slow (micros-ms) time scale dynamics in protein side chains by (¹⁵N) relaxation dispersion NMR spectroscopy: application to Asn and Gln residues in a cavity mutant of T4 lysozyme, *J Am Chem Soc* 123, 967-975.
103. Ishima, R., and Torchia, D. A. (2003) Extending the range of amide proton relaxation dispersion experiments in proteins using a constant-time relaxation-compensated CPMG approach, *J Biomol NMR* 25, 243-248.
104. Vallurupalli, P., Hansen, D. F., Stollar, E., Meirovitch, E., and Kay, L. E. (2007) Measurement of bond vector orientations in invisible excited states of proteins, *Proc Natl Acad Sci U S A* 104, 18473-18477.
105. Tugarinov, V., Kanelis, V., and Kay, L. E. (2006) Isotope labeling strategies for the study of high-molecular-weight proteins by solution NMR spectroscopy, *Nat Protoc* 1, 749-754.
106. Lundstrom, P., Vallurupalli, P., Hansen, D. F., and Kay, L. E. (2009) Isotope labeling methods for studies of excited protein states by relaxation dispersion NMR spectroscopy, *Nat Protoc* 4, 1641-1648.
107. Kovrigin, E. L., and Loria, J. P. (2006) Enzyme dynamics along the reaction coordinate: critical role of a conserved residue, *Biochemistry* 45, 2636-2647.

108. Fraser, J. S., Clarkson, M. W., Degnan, S. C., Erion, R., Kern, D., and Alber, T. (2009) Hidden alternative structures of proline isomerase essential for catalysis, *Nature* 462, 669-673.
109. Vallurupalli, P., and Kay, L. E. (2006) Complementarity of ensemble and single-molecule measures of protein motion: a relaxation dispersion NMR study of an enzyme complex, *Proc Natl Acad Sci U S A* 103, 11910-11915.
110. Beach, H., Cole, R., Gill, M. L., and Loria, J. P. (2005) Conservation of mus-ms enzyme motions in the apo- and substrate-mimicked state, *J Am Chem Soc* 127, 9167-9176.
111. Sprangers, R., Gribun, A., Hwang, P. M., Houry, W. A., and Kay, L. E. (2005) Quantitative NMR spectroscopy of supramolecular complexes: dynamic side pores in ClpP are important for product release, *Proc Natl Acad Sci U S A* 102, 16678-16683.
112. Doucet, N., Watt, E. D., and Loria, J. P. (2009) The flexibility of a distant loop modulates active site motion and product release in ribonuclease A, *Biochemistry* 48, 7160-7168.
113. Korzhnev, D. M., Neudecker, P., Mittermaier, A., Orekhov, V. Y., and Kay, L. E. (2005) Multiple-site exchange in proteins studied with a suite of six NMR relaxation dispersion experiments: an application to the folding of a Fyn SH3 domain mutant, *J Am Chem Soc* 127, 15602-15611.
114. Schanda, P., Brutscher, B., Konrat, R., and Tollinger, M. (2008) Folding of the KIX domain: characterization of the equilibrium analog of a folding intermediate using ¹⁵N/¹³C relaxation dispersion and fast ¹H/²H amide exchange NMR spectroscopy, *J Mol Biol* 380, 726-741.
115. Sugase, K., Dyson, H. J., and Wright, P. E. (2007) Mechanism of coupled folding and binding of an intrinsically disordered protein, *Nature* 447, 1021-1025.
116. Farber, P., Darmawan, H., Sprules, T., and Mittermaier, A. (2010) Analyzing Protein Folding Cooperativity by Differential Scanning Calorimetry and NMR Spectroscopy, *J Am Chem Soc* 132, 6214-6222.
117. Tzeng, S.-R., and Kalodimos, C. G. (2009) Dynamic activation of an allosteric regulatory protein, *Nature* 462, 368-372.
118. Wolf-Watz, M., Thai, V., Henzler-Wildman, K., Hadjipavlou, G., Eisenmesser, E. Z., and Kern, D. (2004) Linkage between dynamics and catalysis in a thermophilic-mesophilic enzyme pair, *Nat Struct Mol Biol* 11, 945-949.
119. Eisenmesser, E. Z., Millet, O., Labeikovsky, W., Korzhnev, D. M., Wolf-Watz, M., Bosco, D. A., Skalicky, J. J., Kay, L. E., and Kern, D. (2005) Intrinsic dynamics of an enzyme underlies catalysis, *Nature* 438, 117-121.
120. Korzhnev, D. M., and Kay, L. E. (2008) Probing invisible, low-populated States of protein molecules by relaxation dispersion NMR spectroscopy: an application to protein folding, *Acc Chem Res* 41, 442-451.
121. Korzhnev, D. M., Neudecker, P., Mittermaier, A., Orekhov, V. Y., and Kay, L. E. (2005) Multiple-Site Exchange in Proteins Studied with a Suite of Six NMR Relaxation Dispersion Experiments: An Application to the Folding of a Fyn SH3 Domain Mutant, *J Am Chem Soc* 127, 15602-15611.
122. Skrynnikov, N. R., Dahlquist, F. W., and Kay, L. E. (2002) Reconstructing NMR Spectra of "Invisible" Excited Protein States Using HSQC and HMQC Experiments, *J Am Chem Soc* 124, 12352-12360.
123. Bouvignies, G., Vallurupalli, P., Hansen, D. F., Correia, B. E., Lange, O., Bah, A., Vernon, R. M., Dahlquist, F. W., Baker, D., and Kay, L. E. (2011) Solution structure of a minor and transiently formed state of a T4 lysozyme mutant, *Nature* 477, 111-114.
124. Jeener, J., Meier, B. H., Bachmann, P., and Ernst, R. R. (1979) Investigation of exchange processes by two-dimensional NMR spectroscopy, *J Chem Phys* 71, 4546-4553.
125. Religa, T. L., Sprangers, R., and Kay, L. E. (2010) Dynamic regulation of archaeal proteasome gate opening as studied by TROSY NMR, *Science* 328, 98-102.

126. Sahu, D., Clore, G. M., and Iwahara, J. (2007) TROSY-Based z-Exchange Spectroscopy: Application to the Determination of the Activation Energy for Intermolecular Protein Translocation between Specific Sites on Different DNA Molecules, *J Am Chem Soc* 129, 13232-13237.
127. Kuloglu, E. S., McCaslin, D. R., Markley, J. L., and Volkman, B. F. (2002) Structural rearrangement of human lymphotactin, a C chemokine, under physiological solution conditions, *J Biol Chem* 277, 17863-17870.
128. Hwang, P. M., Bishop, R. E., and Kay, L. E. (2004) The integral membrane enzyme PagP alternates between two dynamically distinct states, *Proc Natl Acad Sci U S A* 101, 9618-9623.
129. Nikolaev, Y., and Pervushin, K. (2007) NMR Spin State Exchange Spectroscopy Reveals Equilibrium of Two Distinct Conformations of Leucine Zipper GCN4 in Solution, *J Am Chem Soc* 129, 6461-6469.
130. Li, Y., and Palmer, A. G., 3rd. (2009) TROSY-selected ZZ-exchange experiment for characterizing slow chemical exchange in large proteins, *J Biomol NMR* 45, 357-360.
131. Banci, L., Bertini, I., Cavazza, C., Felli, I. C., and Koulougliotis, D. (1998) Probing the backbone dynamics of oxidized and reduced rat microsomal cytochrome b5 via ¹⁵N rotating frame NMR relaxation measurements: biological implications, *Biochemistry* 37, 12320-12330.
132. Bertini, I., Kursula, P., Luchinat, C., Parigi, G., Vahokoski, J., Wilmanns, M., and Yuan, J. (2009) Accurate solution structures of proteins from X-ray data and a minimal set of NMR data: calmodulin-peptide complexes as examples, *J Am Chem Soc* 131, 5134-5144.
133. Hass, M. A., Keizers, P. H., Blok, A., Hiruma, Y., and Ubbink, M. (2010) Validation of a lanthanide tag for the analysis of protein dynamics by paramagnetic NMR spectroscopy, *J Am Chem Soc* 132, 9952-9953.
134. Omura, T., and Sato, R. (1964) The Carbon Monoxide-Binding Pigment of Liver Microsomes. I. Evidence for Its Hemoprotein Nature, *J Biol Chem* 239, 2370-2378.
135. Nelson, D. R., Koymans, L., Kamataki, T., Stegeman, J. J., Feyereisen, R., Waxman, D. J., Waterman, M. R., Gotoh, O., Coon, M. J., Estabrook, R. W., Gunsalus, I. C., and Nebert, D. W. (1996) P450 superfamily: update on new sequences, gene mapping, accession numbers and nomenclature, *Pharmacogenetics* 6, 1-42.
136. McLean, K. J., Sabri, M., Marshall, K. R., Lawson, R. J., Lewis, D. G., Clift, D., Balding, P. R., Dunford, A. J., Warman, A. J., McVey, J. P., Quinn, A. M., Sutcliffe, M. J., Scrutton, N. S., and Munro, A. W. (2005) Biodiversity of cytochrome P450 redox systems, *Biochem Soc Trans* 33, 796-801.
137. Williams, P. A., Cosme, J., Ward, A., Angove, H. C., Matak Vinkovic, D., and Jhoti, H. (2003) Crystal structure of human cytochrome P450 2C9 with bound warfarin, *Nature* 424, 464-468.
138. Lewis, D. F. (1999) Homology modelling of human cytochromes P450 involved in xenobiotic metabolism and rationalization of substrate selectivity, *Exp Toxicol Pathol* 51, 369-374.
139. Ortiz de Montellano, P. R. (2005) *Cytochrome P450: Structure, Mechanism and Biochemistry*, 3 ed., Kluwer Academic/Plenum Publishers, New York.
140. Poulos, T. L., Finzel, B. C., Gunsalus, I. C., Wagner, G. C., and Kraut, J. (1985) The 2.6-A crystal structure of *Pseudomonas putida* cytochrome P-450, *J Biol Chem* 260, 16122-16130.
141. Lee, Y. T., Wilson, R. F., Rupniewski, I., and Goodin, D. B. (2010) P450cam visits an open conformation in the absence of substrate, *Biochemistry* 49, 3412-3419.
142. Ludemann, S. K., Lounnas, V., and Wade, R. C. (2000) How do substrates enter and products exit the buried active site of cytochrome P450cam? 1. Random expulsion molecular dynamics investigation of ligand access channels and mechanisms, *J Mol Biol* 303, 797-811.
143. Winn, P. J., Ludemann, S. K., Gauges, R., Lounnas, V., and Wade, R. C. (2002) Comparison of the dynamics of substrate access channels in three cytochrome

- P450s reveals different opening mechanisms and a novel functional role for a buried arginine, *Proc Natl Acad Sci U S A* 99, 5361-5366.
144. Cojocaru, V., Winn, P. J., and Wade, R. C. (2007) The ins and outs of cytochrome P450s, *Biochim Biophys Acta* 1770, 390-401.
 145. Stoll, S., Lee, Y. T., Zhang, M., Wilson, R. F., Britt, R. D., and Goodin, D. B. (2012) Double electron-electron resonance shows cytochrome P450cam undergoes a conformational change in solution upon binding substrate, *Proc Natl Acad Sci U S A* 109, 12888-12893.
 146. Poulos, T. L., Finzel, B. C., and Howard, A. J. (1987) High-resolution crystal structure of cytochrome P450cam, *J Mol Biol* 195, 687-700.
 147. McHaourab, H. S., Oh, K. J., Fang, C. J., and Hubbell, W. L. (1997) Conformation of T4 lysozyme in solution. Hinge-bending motion and the substrate-induced conformational transition studied by site-directed spin labeling, *Biochemistry* 36, 307-316.
 148. Goto, N. K., Skrynnikov, N. R., Dahlquist, F. W., and Kay, L. E. (2001) What is the average conformation of bacteriophage T4 lysozyme in solution? A domain orientation study using dipolar couplings measured by solution NMR, *J Mol Biol* 308, 745-764.
 149. de Groot, B. L., Hayward, S., van Aalten, D. M., Amadei, A., and Berendsen, H. J. (1998) Domain motions in bacteriophage T4 lysozyme: a comparison between molecular dynamics and crystallographic data, *Proteins* 31, 116-127.
 150. Mulder, F. A., Mittermaier, A., Hon, B., Dahlquist, F. W., and Kay, L. E. (2001) Studying excited states of proteins by NMR spectroscopy, *Nat Struct Biol* 8, 932-935.
 151. Yirdaw, R. B., and McHaourab, H. S. (2012) Direct observation of T4 lysozyme hinge-bending motion by fluorescence correlation spectroscopy, *Biophys J* 103, 1525-1536.
 152. Vetter, I. R., Baase, W. A., Heinz, D. W., Xiong, J. P., Snow, S., and Matthews, B. W. (1996) Protein structural plasticity exemplified by insertion and deletion mutants in T4 lysozyme, *Protein Sci* 5, 2399-2415.
 153. Luderer, R., Takken, F. L., de Wit, P. J., and Joosten, M. H. (2002) *Cladosporium fulvum* overcomes Cf-2-mediated resistance by producing truncated AVR2 elicitor proteins, *Mol Microbiol* 45, 875-884.
 154. Dixon, M. S., Jones, D. A., Keddle, J. S., Thomas, C. M., Harrison, K., and Jones, J. D. (1996) The tomato Cf-2 disease resistance locus comprises two functional genes encoding leucine-rich repeat proteins, *Cell* 84, 451-459.
 155. Kruger, J., Thomas, C. M., Golstein, C., Dixon, M. S., Smoker, M., Tang, S., Mulder, L., and Jones, J. D. (2002) A tomato cysteine protease required for Cf-2-dependent disease resistance and suppression of autonecrosis, *Science* 296, 744-747.
 156. Rooney, H. C., Van't Klooster, J. W., van der Hoorn, R. A., Joosten, M. H., Jones, J. D., and de Wit, P. J. (2005) *Cladosporium Avr2* inhibits tomato Rcr3 protease required for Cf-2-dependent disease resistance, *Science* 308, 1783-1786.
 157. Shabab, M., Shindo, T., Gu, C., Kaschani, F., Pansuriya, T., Chinttha, R., Harzen, A., Colby, T., Kamoun, S., and van der Hoorn, R. A. (2008) Fungal effector protein AVR2 targets diversifying defense-related cys proteases of tomato, *Plant Cell* 20, 1169-1183.
 158. Axtell, M. J., and Staskawicz, B. J. (2003) Initiation of RPS2-specified disease resistance in *Arabidopsis* is coupled to the AvrRpt2-directed elimination of RIN4, *Cell* 112, 369-377.
 159. Berendt, U., Haverkamp, T., Prior, A., and Schwenn, J. D. (1995) Reaction mechanism of thioredoxin: 3'-phospho-adenylylsulfate reductase investigated by site-directed mutagenesis, *Eur J Biochem* 233, 347-356.
 160. Koprivova, A., Meyer, A. J., Schween, G., Herschbach, C., Reski, R., and Kopriva, S. (2002) Functional knockout of the adenosine 5'-phosphosulfate reductase gene in *Physcomitrella patens* revives an old route of sulfate assimilation, *J Biol Chem* 277, 32195-32201.

161. Lillig, C. H., Prior, A., Schwenn, J. D., Aslund, F., Ritz, D., Vlamis-Gardikas, A., and Holmgren, A. (1999) New thioredoxins and glutaredoxins as electron donors of 3'-phosphoadenylylsulfate reductase, *J Biol Chem* 274, 7695-7698.
162. Bick, J. A., Dennis, J. J., Zylstra, G. J., Nowack, J., and Leustek, T. (2000) Identification of a new class of 5'-adenylylsulfate (APS) reductases from sulfate-assimilating bacteria, *J Bacteriol* 182, 135-142.
163. Carroll, K. S., Gao, H., Chen, H., Stout, C. D., Leary, J. A., and Bertozzi, C. R. (2005) A conserved mechanism for sulfonucleotide reduction, *PLoS Biol* 3, e250.
164. Kim, S. K., Rahman, A., Bick, J. A., Conover, R. C., Johnson, M. K., Mason, J. T., Hirasawa, M., Leustek, T., and Knaff, D. B. (2004) Properties of the cysteine residues and iron-sulfur cluster of the assimilatory 5'-adenylyl sulfate reductase from *Pseudomonas aeruginosa*, *Biochemistry* 43, 13478-13486.
165. Kopriva, S., Buchert, T., Fritz, G., Suter, M., Benda, R., Schunemann, V., Koprivova, A., Schurmann, P., Trautwein, A. X., Kroneck, P. M., and Brunold, C. (2002) The presence of an iron-sulfur cluster in adenosine 5'-phosphosulfate reductase separates organisms utilizing adenosine 5'-phosphosulfate and phosphoadenosine 5'-phosphosulfate for sulfate assimilation, *J Biol Chem* 277, 21786-21791.
166. Berndt, C., Lillig, C. H., Wollenberg, M., Bill, E., Mansilla, M. C., de Mendoza, D., Seidler, A., and Schwenn, J. D. (2004) Characterization and reconstitution of a 4Fe-4S adenylyl sulfate/phosphoadenylyl sulfate reductase from *Bacillus subtilis*, *J Biol Chem* 279, 7850-7855.
167. Gao, Y., Schofield, O. M., and Leustek, T. (2000) Characterization of sulfate assimilation in marine algae focusing on the enzyme 5'-adenylylsulfate reductase, *Plant Physiol* 123, 1087-1096.
168. Kim, S. K., Rahman, A., Conover, R. C., Johnson, M. K., Mason, J. T., Gomes, V., Hirasawa, M., Moore, M. L., Leustek, T., and Knaff, D. B. (2006) Properties of the cysteine residues and the iron-sulfur cluster of the assimilatory 5'-adenylyl sulfate reductase from *Enteromorpha intestinalis*, *Biochemistry* 45, 5010-5018.
169. Bick, J. A., Aslund, F., Chen, Y., and Leustek, T. (1998) Glutaredoxin function for the carboxyl-terminal domain of the plant-type 5'-adenylylsulfate reductase, *Proc Natl Acad Sci U S A* 95, 8404-8409.
170. Kim, S. K., Gomes, V., Gao, Y., Chandramouli, K., Johnson, M. K., Knaff, D. B., and Leustek, T. (2007) The two-domain structure of 5'-adenylylsulfate (APS) reductase from *Enteromorpha intestinalis* is a requirement for efficient APS reductase activity, *Biochemistry* 46, 591-601.
171. Sun, Q. A., Su, D., Novoselov, S. V., Carlson, B. A., Hatfield, D. L., and Gladyshev, V. N. (2005) Reaction mechanism and regulation of mammalian thioredoxin/glutathione reductase, *Biochemistry* 44, 14528-14537.
172. Rouhier, N., Gama, F., Wingsle, G., Gelhaye, E., Gans, P., and Jacquot, J. P. (2006) Engineering functional artificial hybrid proteins between poplar peroxiredoxin II and glutaredoxin or thioredoxin, *Biochem Biophys Res Commun* 341, 1300-1308.
173. Li, X., Nield, J., Hayman, D., and Langridge, P. (1995) Thioredoxin activity in the C terminus of Phalaris S protein, *Plant J* 8, 133-138.
174. Delaglio, F., Grzesiek, S., Vuister, G. W., Zhu, G., Pfeifer, J., and Bax, A. (1995) NMRPipe: a multidimensional spectral processing system based on UNIX pipes, *J Biomol NMR* 6, 277-293.
175. Vranken, W. F., Boucher, W., Stevens, T. J., Fogh, R. H., Pajon, A., Llinas, M., Ulrich, E. L., Markley, J. L., Ionides, J., and Laue, E. D. (2005) The CCPN data model for NMR spectroscopy: development of a software pipeline, *Proteins* 59, 687-696.
176. Shen, Y., and Bax, A. (2012) Identification of helix capping and b-turn motifs from NMR chemical shifts, *J Biomol NMR* 52, 211-232.
177. LeMaster, D. M. (1994) Isotope labeling in solution protein assignment and structural analysis, *Progress in Nuclear Magnetic Resonance Spectroscopy* 26, Part 4, 371-419.

178. Sattler, M., and Fesik, S. W. (1996) Use of deuterium labeling in NMR: overcoming a sizeable problem, *Structure* 4, 1245-1249.
179. Otten, R., Chu, B., Krewulak, K. D., Vogel, H. J., and Mulder, F. A. (2010) Comprehensive and cost-effective NMR spectroscopy of methyl groups in large proteins, *J Am Chem Soc* 132, 2952-2960.
180. Lee, Y. T., Glazer, E. C., Wilson, R. F., Stout, C. D., and Goodin, D. B. (2011) Three clusters of conformational states in p450cam reveal a multistep pathway for closing of the substrate access channel, *Biochemistry* 50, 693-703.
181. Gunsalus, I. C., and Sligar, S. G. (1976) Redox regulation of cytochrome P450cam mixed function oxidation by putidaredoxin and camphor ligation, *Biochimie* 58, 143-147.
182. Fedorov, R., Ghosh, D. K., and Schlichting, I. (2003) Crystal structures of cyanide complexes of P450cam and the oxygenase domain of inducible nitric oxide synthase, structural models of the short-lived oxygen complexes, *Archives of Biochemistry and Biophysics* 409, 25-31.
183. Poulos, T. L., and Howard, A. J. (1987) Crystal structures of metyrapone- and phenylimidazole-inhibited complexes of cytochrome P-450cam, *Biochemistry* 26, 8165-8174.
184. Grzesiek, S., Bax, A., Clore, G. M., Gronenborn, A. M., Hu, J. S., Kaufman, J., Palmer, I., Stahl, S. J., and Wingfield, P. T. (1996) The solution structure of HIV-1 Nef reveals an unexpected fold and permits delineation of the binding surface for the SH3 domain of Hck tyrosine protein kinase, *Nat Struct Biol* 3, 340-345.
185. Crull, G. B., Kennington, J. W., Garber, A. R., Ellis, P. D., and Dawson, J. H. (1989) ¹⁹F nuclear magnetic resonance as a probe of the spatial relationship between the heme iron of cytochrome P-450 and its substrate, *J Biol Chem* 264, 2649-2655.
186. Thomma, B. P., HP, V. A. N. E., Crous, P. W., and PJ, D. E. W. (2005) *Cladosporium fulvum* (syn. *Passalora fulva*), a highly specialized plant pathogen as a model for functional studies on plant pathogenic Mycosphaerellaceae, *Mol Plant Pathol* 6, 379-393.
187. Horger, A. C., and van der Hoorn, R. A. (2013) The structural basis of specific protease-inhibitor interactions at the plant-pathogen interface, *Curr Opin Struct Biol*.
188. Van't Klooster, J. W., Van der Kamp, M. W., Vervoort, J., Beekwilder, J., Boeren, S., Joosten, M. H., Thomma, B. P., and De Wit, P. J. (2011) Affinity of Avr2 for tomato cysteine protease Rcr3 correlates with the Avr2-triggered Cf-2-mediated hypersensitive response, *Mol Plant Pathol* 12, 21-30.
189. Nederveen, A. J., Doreleijers, J. F., Vranken, W., Miller, Z., Spronk, C. A., Nabuurs, S. B., Guntert, P., Livny, M., Markley, J. L., Nilges, M., Ulrich, E. L., Kaptein, R., and Bonvin, A. M. (2005) RECOORD: a recalculated coordinate database of 500+ proteins from the PDB using restraints from the BioMagResBank, *Proteins* 59, 662-672.
190. Doreleijers, J. F., Sousa da Silva, A. W., Krieger, E., Nabuurs, S. B., Spronk, C. A., Stevens, T. J., Vranken, W. F., Vriend, G., and Vuister, G. W. (2012) CING: an integrated residue-based structure validation program suite, *J Biomol NMR* 54, 267-283.
191. Shen, Y., Delaglio, F., Cornilescu, G., and Bax, A. (2009) TALOS+: a hybrid method for predicting protein backbone torsion angles from NMR chemical shifts, *J Biomol NMR* 44, 213-223.
192. Brunger, A. T., Adams, P. D., Clore, G. M., DeLano, W. L., Gros, P., Grosse-Kunstleve, R. W., Jiang, J. S., Kuszewski, J., Nilges, M., Pannu, N. S., Read, R. J., Rice, L. M., Simonson, T., and Warren, G. L. (1998) Crystallography & NMR system: A new software suite for macromolecular structure determination, *Acta Crystallogr D Biol Crystallogr* 54, 905-921.
193. Brunger, A. T. (2007) Version 1.2 of the Crystallography and NMR system, *Nat Protoc* 2, 2728-2733.

194. Laskowski, R. A., Rullmannn, J. A., MacArthur, M. W., Kaptein, R., and Thornton, J. M. (1996) AQUA and PROCHECK-NMR: programs for checking the quality of protein structures solved by NMR, *J Biomol NMR* 8, 477-486.
195. Tripathi, S., Li, H., and Poulos, T. L. (2013) Structural basis for effector control and redox partner recognition in cytochrome P450, *Science* 340, 1227-1230.
196. Hiruma, Y., Hass, M. A., Kikui, Y., Liu, W. M., Olmez, B., Skinner, S. P., Blok, A., Kloosterman, A., Koteishi, H., Lohr, F., Schwalbe, H., Nojiri, M., and Ubbink, M. (2013) The Structure of the Cytochrome P450cam-Putidaredoxin Complex Determined by Paramagnetic NMR Spectroscopy and Crystallography, *J Mol Biol.*
197. Sjodin, T., Christian, J. F., Macdonald, I. D., Davydov, R., Unno, M., Sligar, S. G., Hoffman, B. M., and Champion, P. M. (2001) Resonance Raman and EPR investigations of the D251N oxycytochrome P450cam/putidaredoxin complex, *Biochemistry* 40, 6852-6859.
198. Myers, W. K., Lee, Y. T., Britt, R. D., and Goodin, D. B. (2013) The conformation of p450cam in complex with putidaredoxin is dependent on oxidation state, *J Am Chem Soc* 135, 11732-11735.
199. Franke, A., Hartmann, E., Schlichting, I., and van Eldik, R. (2012) A complete volume profile for the reversible binding of camphor to cytochrome P450(cam), *J Biol Inorg Chem* 17, 447-463.
200. Hays, A. M., Dunn, A. R., Chiu, R., Gray, H. B., Stout, C. D., and Goodin, D. B. (2004) Conformational states of cytochrome P450cam revealed by trapping of synthetic molecular wires, *J Mol Biol* 344, 455-469.
201. Dunn, A. R., Dmochowski, I. J., Bilwes, A. M., Gray, H. B., and Crane, B. R. (2001) Probing the open state of cytochrome P450cam with ruthenium-linker substrates, *Proc Natl Acad Sci U S A* 98, 12420-12425.
202. Liu, W.-M., Keizers, P. H. J., Hass, M. A. S., Blok, A., Timmer, M., Sarris, A. J. C., Overhand, M., and Ubbink, M. (2012) A pH-Sensitive, Colorful, Lanthanide-Chelating Paramagnetic NMR Probe, *Journal of the American Chemical Society* 134, 17306-17313.
203. Riek, R., Fiaux, J., Bertelsen, E. B., Horwich, A. L., and Wuthrich, K. (2002) Solution NMR techniques for large molecular and supramolecular structures, *J Am Chem Soc* 124, 12144-12153.
204. Salzmann, M., Pervushin, K., Wider, G., Senn, H., and Wuthrich, K. (1998) TROSY in triple-resonance experiments: new perspectives for sequential NMR assignment of large proteins, *Proc Natl Acad Sci U S A* 95, 13585-13590.
205. Fiaux, J., Bertelsen, E. B., Horwich, A. L., and Wuthrich, K. (2002) NMR analysis of a 900K GroEL GroES complex, *Nature* 418, 207-211.
206. Kay, L. E. (2011) Solution NMR spectroscopy of supra-molecular systems, why bother? A methyl-TROSY view, *J Magn Reson* 210, 159-170.
207. Amero, C., Asuncion Dura, M., Noirclerc-Savoye, M., Perollier, A., Gallet, B., Plevin, M., Vernet, T., Franzetti, B., and Boisbouvier, J. (2011) A systematic mutagenesis-driven strategy for site-resolved NMR studies of supramolecular assemblies, *J Biomol NMR* 50, 229-236.
208. Sprangers, R., Li, X., Mao, X., Rubinstein, J. L., Schimmer, A. D., and Kay, L. E. (2008) TROSY-based NMR evidence for a novel class of 20S proteasome inhibitors, *Biochemistry* 47, 6727-6734.
209. Keizers, P. H., Mersinli, B., Reinle, W., Donauer, J., Hiruma, Y., Hannemann, F., Overhand, M., Bernhardt, R., and Ubbink, M. (2010) A solution model of the complex formed by adrenodoxin and adrenodoxin reductase determined by paramagnetic NMR spectroscopy, *Biochemistry* 49, 6846-6855.
210. Luchinat, C., Parigi, G., Ravera, E., and Rinaldelli, M. (2012) Solid-State NMR Crystallography through Paramagnetic Restraints, *J Am Chem Soc* 134, 5006-5009.
211. Bertini, I., Bhaumik, A., De Paëpe, G., Griffin, R. G., Lelli, M., Lewandowski, J. z. R., and Luchinat, C. (2009) High-Resolution Solid-State NMR Structure of a 17.6 kDa Protein, *J Am Chem Soc* 132, 1032-1040.

212. Keizers, P. H., Desreux, J. F., Overhand, M., and Ubbink, M. (2007) Increased paramagnetic effect of a lanthanide protein probe by two-point attachment, *J Am Chem Soc* 129, 9292-9293.
213. Jia, X., Yagi, H., Su, X. C., Stanton-Cook, M., Huber, T., and Otting, G. (2011) Engineering [Ln(DPA)₃] 3- binding sites in proteins: a widely applicable method for tagging proteins with lanthanide ions, *J Biomol NMR* 50, 411-420.
214. Bertini, I., Luchinat, C., and Parigi, G. (2002) Magnetic susceptibility in paramagnetic NMR, *Prog Nucl Mag Res Sp* 40, 249-273.
215. Koehler, J., and Meiler, J. (2011) Expanding the utility of NMR restraints with paramagnetic compounds: background and practical aspects, *Prog Nucl Magn Reson Spectrosc* 59, 360-389.
216. Kraft, D. (1988) A software package for sequential quadratic programming, in *Tech. Rep. DFVLR-FB 88-28*, Institute for Flight Mechanics, Koln.
217. Kuhn, H. W. (1955) The Hungarian method for the assignment problem, *Nav Res Logist Q* 2, 83-97.
218. Hamelryck, T., and Manderick, B. (2003) PDB file parser and structure class implemented in Python, *Bioinformatics* 19, 2308-2310.
219. Cock, P. J., Antao, T., Chang, J. T., Chapman, B. A., Cox, C. J., Dalke, A., Friedberg, I., Hamelryck, T., Kauff, F., Wilczynski, B., and de Hoon, M. J. (2009) Biopython: freely available Python tools for computational molecular biology and bioinformatics, *Bioinformatics* 25, 1422-1423.
220. Prudencio, M., Rohovec, J., Peters, J. A., Tocheva, E., Boulanger, M. J., Murphy, M. E., Hupkes, H. J., Kusters, W., Impagliazzo, A., and Ubbink, M. (2004) A caged lanthanide complex as a paramagnetic shift agent for protein NMR, *Chemistry* 10, 3252-3260.
221. Schlichting, I., Berendzen, J., Chu, K., Stock, A. M., Maves, S. A., Benson, D. E., Sweet, R. M., Ringe, D., Petsko, G. A., and Sligar, S. G. (2000) The catalytic pathway of cytochrome p450cam at atomic resolution, *Science* 287, 1615-1622.
222. Eichmuller, C., and Skrynnikov, N. R. (2007) Observation of microsecond time-scale protein dynamics in the presence of Ln³⁺ ions: application to the N-terminal domain of cardiac troponin C, *J Biomol NMR* 37, 79-95.
223. McIntosh, L. P., Wand, A. J., Lowry, D. F., Redfield, A. G., and Dahlquist, F. W. (1990) Assignment of the backbone ¹H and ¹⁵N NMR resonances of bacteriophage T4 lysozyme, *Biochemistry* 29, 6341-6362.
224. Skinner, S. P., Moshev, M., Hass, M. A., and Ubbink, M. (2013) PARAssign--paramagnetic NMR assignments of protein nuclei on the basis of pseudocontact shifts, *J Biomol NMR* 55, 379-389.
225. Bardiaux, B., Malliavin, T., and Nilges, M. (2012) ARIA for solution and solid-state NMR, *Methods Mol Biol* 831, 453-483.
226. Bouvignies, G., and Kay, L. E. (2012) A 2D (1)(3)C-CEST experiment for studying slowly exchanging protein systems using methyl probes: an application to protein folding, *J Biomol NMR* 53, 303-310.