

## IV. Conclusions et perspectives

Le fort potentiel des complexes pour la préparation de dérivés présentant un fort pouvoir de phasage, tant avec un générateur de laboratoire qu'avec le rayonnement synchrotron, a été confirmé.

Dans de nombreux cas, des cristaux dérivés obtenus grâce à l'utilisation des complexes ont mené à des cartes de densité électroniques expérimentales d'excellente qualité. Ces cartes permettent non seulement de construire aisément le modèle de la protéine, mais également de placer d'éventuels ligands (voir par exemple le cas de l'inhibiteur  $\delta$ -azaxanthine dans la protéine urate oxydase, figure 2.3.2 du § II.3.6).

Outre leurs propriétés de fixation dans les cristaux et leur forte diffusion anormale, l'avantage des complexes est leur facilité d'utilisation et leur bonne tolérance par les protéines. La complémentarité du mode fixation des différents complexes sur les protéines fait que, en testant plusieurs d'entre eux, on augmente les chances d'obtenir un dérivé avec un taux de fixation élevé.

Les résultats des affinements du mode de fixation observé pour différentes combinaisons de complexe et de protéine permettent, dans certains cas, d'orienter le choix du complexe à utiliser avec une nouvelle protéine. Toutefois, ces résultats ne permettent pas, dans le cas général, de prédire de manière certaine la fixation d'un complexe sur une nouvelle protéine.

Dans les cas étudiés, les méthodes alternatives à la cristallographie qui permettent de caractériser l'interaction d'une protéine avec un ligand n'ont pas permis d'estimer le taux de fixation d'un complexe avec la protéine utilisée.

Néanmoins, l'utilisation des complexes par d'autres groupes de cristallographie biologique, comme alternative à des méthodes établies de préparation de dérivés lourds, serait tout à fait souhaitable. Dans ce dessein, il faudrait rendre facilement accessible l'ensemble des complexes par des équipes intéressées.

Ceci n'est pas le cas actuellement (voir § I.5.2)\*\*. Il serait souhaitable, que l'ensemble des complexes soit commercialisé sous forme de crible qui permette de tester les différents complexes pour l'obtention d'un dérivé lourd. Dans ce dessein, le fournisseur de matériel de cristallisation Hampton Research a contacté notre équipe dans le passé. Les négociations qui ont suivi entre les entreprises Hampton Research et Bracco Imaging n'ont pas abouti à la commercialisation d'un tel crible. La levée, récente, d'un certain nombre de brevets protégeant la synthèse des complexes pourrait permettre la relance d'une opération similaire.

Un tel crible, en combinaison avec des procédures de traitements de données de diffraction rapides, automatisés qui permettent l'estimation du pouvoir de phasage d'un dérivé pendant l'acquisition des données de diffraction (Panjikar *et al.*, 2005), permettrait l'obtention efficace de dérivés avec un bon pouvoir de phasage, leur vérification et, le cas échéant, la mise à l'écart de dérivés qui ne permettent pas d'obtenir les phases.

Les résultats des études sur l'obtention des dérivés et sur les modes de fixation des complexes analysés à partir des structures affinées, pourraient aider à concevoir de nouveaux ligands d'atomes lourds, optimisés pour la fixation dans les cristaux de protéines.

## Références

- J. Abrahams (1997). "Bias reduction in phase refinement by modified interference functions: introducing the gamma correction." *Acta Cryst. D***52**, 43-48.
- S. Aime, E. Gianolio, E. Terreno, G. Giovenzana, R. Pagliarin, M. Sisti, G. Palmisano, M. Botta, M. Lowe & D. Parker (2000). "Ternary Gd(III)L-HSA adducts: evidence for the replacement of inner-sphere water molecules by coordinating groups of the protein. Implications for the design of contrast agents for MRI." *JBIC*, **5**, 488-497.
- P. Anelli, V. Balzani, L. Prodi & F. Uggeri (1991). "Luminescence properties of Eu<sup>3+</sup> complexes of highly polydentate ligands." *Gazetta Chimica Italiana*, **121**, 359-364.
- A. Bianchi, L. Calabi, F. Corana, S. Fontana, P. Losi, A. Maiocchi, L. Paleari & B. Valtancoli (2000). "Thermodynamic and structural properties of Gd(III) complexes with polyamino-polycarboxylic ligands: basic compounds for the development of MRI contrast agents." *Coord. Chem. Rev.* **204**, 309-393.
- K. Binnemans & C. Görller-Walrand (1995). "On the color of the trivalent lanthanide ions." *Chemical Physics Letters*, **235**, 163-174.
- D. Blow & F. Crick (1959). *Acta Cryst.* **12**, 794-802.
- T. Boggon & L. Shapiro (2000). "Screening for phasing atoms in protein crystallography." *Structure*, **8**, R143-R149.
- F. Bonneté, D. Vivarès, C. Robert & N. Colloc'h (2001). "Interactions in solution and crystallization of *Aspergillus flavus* urate oxidase" *J. Crystal Growth* **232**, 330-339.
- D. Bourgeois, X. Vernede, V. Adams, E. Fioravanti & T. Ursby (2002). "A microspectrophotometer for UV-visible absorption and fluorescence studies of protein crystals." *J. Appl. Cryst.* **35**, 319-326.
- G. Bricogne (1988). "A Bayesian statistical theory of the phase problem. I. A multichannel maximum-entropy formalism for constructing generalized joint probability distributions of structure factors." *Acta Cryst. A***44**, 517-545.
- G. Bricogne, C. Vonrhein, C. Flensburg, M. Schiltz & W. Paciorek (2003). "Generation, representation and flow of phase information in structure determination: recent developments in and around SHARP 2.0." *Acta Cryst. D***59**, 2023-2030.
- D. Brooks (2005). <http://dwb.unl.edu>.
- M. Brown, R. Peierls & E. Stern (1997). "White lines in x-ray absorption." *Physical Review B*. **15**, **2**, 738-744.
- A. Brünger (1992). "Free R value: a novel statistical quantity for assessing the accuracy of crystal structures." *Nature* **355**, 472-475

A. Brünger, F. Temple-Burling, P. Grost, R. Grosse-Kunstleve, J. Jiang, J. Kuszewski, M. Nilges, N. Pannu, R. Read, L. Rice, T. Simonson & G. Warren (1998). "Crystallography & NMR System: A new software suite for macromolecular structure determination." *Acta Cryst.* **D54**, 905-921.

J. Bünzli & J. Pfefferle (1994). "Bovine  $\alpha$ -Lactalbumin: Identification of two metal-ion-binding sites using the europium (III) luminescent probe." *Helv. Chim. Acta*, **77**, 323.

W. Burmeister (2000). "Structural changes in a cryo-cooled protein crystal owing to radiation damage." *Acta Cryst.* **D56**, 328-341.

H. Carrell, H. Hoier & J. Glusker (1994). "Modes of binding substrates and their analogs to the enzyme D-xylose isomerase." *Acta Cryst.* **D50**, 113-123.

C. Carubelli, A. Massabni & S. Leite (1997). "Study of the binding of  $\text{Eu}^{3+}$  and  $\text{Tb}^{3+}$  to L-phenylalanine and L-tryptophan." *J. Braz. Chem. Soc.*, **8**, 597-602.

C. Chang, L. Francesconi, M. Malley, K. Kumar, J. Gougoutas, M. Tweedle, D. Lee & L. Wilson (1993). "Synthesis, Characterization, and Crystal Structures of  $\text{M}(\text{DO3A})$  ( $\text{M} = \text{Fe}, \text{Gd}$ ) and  $\text{Na}[\text{M}(\text{DOTA})]$  ( $\text{M} = \text{Fe}, \text{Y}, \text{Gd}$ )." *Inorg. Chem.* **32**, 3501-3508.

C. Charron, R. Giegé & B. Lorber (2004). "Structure of thaumatin in a hexagonal space group: comparison of packing contacts in four crystal lattices." *Acta Cryst.* **D60**, 83-89.

M.-L. Chesne (2002). "Utilisation de la diffusion anormale de l'uranium pour déterminer la structure de macromolécules biologiques - des rayons X durs aux rayons X mous." *Thèse de doctorat, Université Joseph Fourier, Grenoble*.

Collaborative Computational Project, Number 4 (1994). *Acta Cryst.* **D50**, 760-763.

K. Cowtan & P. Main (1996). "Phase combination and cross validation in iterated density-modification calculations" *Acta Cryst.* **D52**, 43-48.

Z. Dauter (2002). "New approaches to high-throughput phasing." *Current opinion in Structural Biology*, **12**:674-678.

Z. Dauter, M. Dauter & E. Dodson (2002). "Jolly SAD." *Acta Cryst* **D58**, 494-506.

Z. Dauter & R. Nagem (2002). "Direct way to anomalous scatterers." *Z. Kristallogr.* **217**, 694-702

W. DeLano (2002). "The PyMOL Molecular Graphics System on World Wide Web." <http://www.pymol.org>

J.-P. Dubost, J.-M. Leger, M.-H. Langlois, D. Meyer & M. Schaefer (1991). "Structure d'un agent de contraste utilisé en imagerie de résonance magnétique le complexe DOTA Gd  $\text{C}_{16}\text{H}_{24}\text{N}_4\text{O}_8\text{NaGd}, 5\text{H}_2\text{O}$ ." *C. R. Acad. Sci. Paris*, **t. 312, Série II**, 349-354.

- L. Ehnebom & B. Pedersen (1992). "Molecular and Crystal Structure of a Lanthanide Complex, Dy-DTPA-BMA Hydrate." *Acta Chemica Scandinavica* **46** 126-130.
- M. Elbanowski & B. Makowska (1996). "The lanthanides as luminescent probes in investigations of biochemical systems." *Journal of Photochemistry and Photobiology A: Chemistry*, **99**, 85-92.
- E. Ennifar, P. Carpentier, J.-L. Ferrer, P. Walter, P. Dumas (2002). "X-ray-induced debromination of nucleic acids at the Br K absorption edge and implications for MAD phasing." *Acta Cryst.* **D58**, 1262-1268.
- G. Evans & G. Bricogne (2002). "Triiodide derivatization and combinatorial counter-ion replacement: two methods for enhancing phasing signal using laboratory Cu K $\alpha$  X-ray equipment" *Acta Cryst.* **D58**, 976-991.
- G. Evans & R. Pettifer (2001). "CHOOCH: a program for deriving anomalous-scattering factors from X-ray fluorescence spectra." *J. Appl. Cryst.* **34**, 82-86.
- A. Feig, M. Panek, W. Horrocks & O. Uhlenbeck (1999). "Probing the binding of Tb(III) and Eu(III) to the hammerhead ribozyme using luminescence spectroscopy." *Chemistry & Biology*, **6**, 801-810.
- E. Garcia, J. Garcia, P. Garcia, A. Arraras, J. Sanchez-Puelles & R. Lopez (1988). "Molecular evolution of lytic enzymes of *Streptococcus pneumoniae* and its bacteriophages." *Proc Natl Acad Sci U S A*, **85(3)**, 914-918.
- E. Garman & J. Murray (2003). "Heavy-atom derivatization." *Acta Cryst.* **D59**, 1903-1913.
- E. Garman & C. Nave (2002). "Radiation damage to crystalline biological molecules: current view." *J. Synchrotron Rad.* **9**, 327-328.
- É. Girard (2001). "Possibilités nouvelles d'études structurales de macromolécules biologiques : utilisation de la diffusion anormale avec des complexes de lanthanides." *Thèse de doctorat, Université Joseph Fourier, Grenoble*.
- É. Girard, P. Anelli, J. Vicat & R. Kahn (2003c). "High-phasing-power lanthanide derivatives: taking advantage of ytterbium and lutetium for optimized anomalous diffraction experiments using synchrotron radiation." *Acta Cryst.* **D59**, 1877-1880.
- É. Girard, L. Chantalat, J. Vicat & R. Kahn (2002). "Gd-HPDO3A, a complex to obtain high-phasing-power heavy-atom derivatives for SAD and MAD experiments: results with tetragonal hen egg-white lysozyme." *Acta Cryst.* **D58**, 1-9.
- É. Girard, M. Stelter, P. L. Anelli, J. Vicat & R. Kahn (2003a). "A new class of gadolinium complexes employed to obtain high-phasing-power heavy-atom derivatives: results from SAD experiments with hen egg-white lysozyme and urate oxidase *Aspergillus flavus*." *Acta Cryst.* **D59**, 118-126.

É. Girard, M. Stelter, J. Vicat & R. Kahn (2003b). "A new class of lanthanide complexes to obtain high-phasing-power heavy-atom derivatives for macromolecular crystallography" *Acta Cryst.* **D59**, 1914–1922.

S. Gras, B. Fernandez, V. Chaumont, P. Carpentier, J. Armengaud & D. Housset (2005). "Expression, purification, crystallization and preliminary crystallographic analysis of the PAB0955 gene product." *Acta Cryst.* **F61**, 208-211.

D. Green, V. Ingram & M. Perutz (1954). "The structure of haemoglobin IV. Sign of determination by isomorphous replacement method." *Proc. R. Soc. London SER. A* **225**, 287-307.

[www.hamptonresearch.com/support/guides/7100G.pdf](http://www.hamptonresearch.com/support/guides/7100G.pdf)

W. Hendrickson (1999). "Maturation of MAD phasing for the determination of macromolecular structures." *J. Synchrotron Rad.* **6**, 845-851.

W. Hendrickson, J. Horton & D. LeMaster (1990). "Selenomethionyl proteins produced for analysis by multiwavelength anomalous diffraction (MAD): a vehicle for direct determination of three-dimensional structure." *EMBO J.*, **9**, 1665-1672.

W. Hendrickson & E. Lattman (1970). "Representation of phase probability distributions for simplified combination of independent phase information." *Acta Cryst.* **B26**, 136-143.

W. Hendrickson & C. Ogata (1997). "Phase determination from multiwavelength anomalous diffraction measurements." *Methods Enzymol.* **276**, 494-523.

W. Hendrickson & M. Teeter (1981). "Structure of the Hydrophobic Protein Crambin Determined Directly from Anomalous Scattering of Sulfur." *Nature*, **290**, 107-113.

J. Hermoso, L. Lagartera, A. González, M. Stelter, P. García, M. Martínez-Ripoll, J. García & M. Menéndez (2005). "Insights into pneumococcal pathogenesis from the crystal structure of the modular teichoic acid phosphorylcholine esterase Pce." *Nature Structural & Molecular Biology* **12**, 533-538.

W. Horrocks (1993). "Luminescence spectroscopy." *Methods Enzymol.*, **226**, 495-538.

W. Horrocks & M. Albin (1991). "Lanthanide Ion Luminescence in Coordination Chemistry and Biochemistry." *Prog. Inorg. Chem.*, **31**, 1-104.

W. Horrocks & W. Collier (1981). "Lanthanide ion luminescence probes. Measurement of distance between intrinsic protein fluorophores and bound metal ions: Quantitation of energy transfer between tryptophan and terbium (III) or europium (III) in the calcium-binding protein parvalbumin." *J. Am. Chem. Soc.*, **103**, 2856-2862.

J. Ibers, D. Templeton, B. Vainshtein, G. Bacon & K. Lonsdale (1985). "Atomic Scattering Factors." in *International Tables for X-ray Crystallography, III, Physical and Chemical tables*. Dordrecht (Holland): D. Reidel Publishing Company for International Union of Crystallography, 201-246.

S. Jeudy, M. Stelter, B. Coutard, R. Kahn & C. Abergel (2005). "Preliminary crystallographic analysis of the Escherichia coli YeaZ protein using the anomalous signal of a gadolinium derivative." *Acta Cryst.* **F61**, 848-851.

T. Jones, J. Zou, S. Cowan & M. Kjeldgaard (1991). "Improved methods for binding protein models in electron density maps and the location of errors in these models." *Acta Cryst.* **A47**, 110-119.

W. Kabsch (1993). "Automatic processing of rotation diffraction data from crystals of initially unknown symmetry and cell constants." *J. Appl. Cryst.* **26**, 795-800.

R. Kahn, P. Carpentier, C. Berthet-Colominas, M. Capitan, M.-L. Chesne, E. Fanchon, S. Lequien, D. Thiaudière, J. Vicat, P. Zielinski & H. Stuhmann (2000). "Feasibility and review of anomalous X-ray diffraction at long wavelengths in materials research and protein crystallography." *J. Synchrotron Rad.* **7**, 131-138.

R. Kahn, R. Fourme, R. Bosshard, M. Chiadmi, J.L. Risler, O. Dideberg, & J.P. Wery (1985). "Crystal structure study of Opsanus tau parvalbumin by multiwavelength anomalous diffraction." *FEBS lett.* **179(1)**, 133-137.

J. Karle & H. Hauptman (1956). "A theory of phase determination for the four types of non-centrosymmetric space groups 1P222, 2P22, 3P<sub>1</sub>2, 3P<sub>2</sub>2." *Acta Cryst.* **9**: 635-651.

G. Kleywegt & T. Jones (1998). "Databases in protein crystallography." *Acta Cryst* **D54**, 1119-1131.

T.-P. Ko, J. Day, A. Greenwood & A. McPherson (1994). "Structures of three crystal forms of the sweet protein thaumatin." *Acta Cryst.* **D50**, 813-825.

H. Kramers (1927). "La diffusion de la lumière par les atomes." *Atti. Congr. Int. Phis. Como* **2**: 545-557.

R. de Kronig (1926). "On the theory of dispersion of X-rays." *J. Opt. Soc. Am.* **12**: 547-557.

K. Kumar, C. Chang, L. Francesconi, D. Dischino, M. Malley, J. Gougoutas & M. Tweedle (1994). "Synthesis, Stability, and Structure of Gadolinium(III) and Yttrium(III) Macrocyclic Poly(amino carboxylates)." *Inorg. Chem.*, **33**, 3567-3575.

E. de La Fortelle & G. Bricogne (1997). "Maximum-likelihood heavy-atom parameter refinement for multiple isomorphous replacement and multiwavelength anomalous diffraction methods." *Methods Enzymol.* **276**, 472-494.

L. Lagartera, A. González, M. Stelter, P. García, R. Kahn, M. Menéndez & J. Hermoso (2005). "Crystallization and preliminary X-ray diffraction studies of the pneumococcal teichoic acid phosphorylcholine esterase Pce." *Acta Cryst.* **F61**, 221-224.

V. Lamzin, A. Perrakis & K. Wilson (2001). "The ARP/WARP suite for automated construction and refinement of protein models." In *Int. Tables for Crystallography. Vol. F: Crystallography of biological macromolecules* (Rossmann, M.G. & Arnold, E. eds.), Dordrecht, Kluwer Academic Publishers, The Netherlands, 720-722.

A. Leslie, E. Duke, D. Brown, J. Naismith, C. Nave, S. Phillips, D. Stuart, A. Thompson, M. Walsh & G. Evans (2004). "Diamond Beamline Proposal 044 - A Tuneable Beamline for Macromolecular Crystallography Optimised for Long Wavelengths (1.5-2.5Å)." Doc. No: BLS-044-PRO-0003.

P. Machin (1985). Editor. *Molecular Replacement*. Proceedings of the Daresbury Study Weekend, 15-16 Feb. 1985. SERC Daresbury Laboratory, Warrington, England.

A. McPherson (2001). "A comparison of salts for the crystallization of macromolecules." *Protein Science*, **10**:418-422.

G. Mie (1908). "Beiträge zur Optik trüber Medien, speziell kolloidaler Metallösungen." *Ann. Phys. (Leipzig)* **25**: 377–452 (Eq. 98).

J. Müller & U. Heinemann (2005). "Use of anomalous scattering for solving the phase problem in protein crystallography." dans : *Encyclopedic Reference of Genomics and Proteomics* (Oschkinat, H. & Schmieder, P. eds) Springer, Heidelberg, sous presse.

J. Murray, E. Garman & R. Ravelli (2004). "X-ray absorption by macromolecular crystals: the effects of wavelength and crystal composition on absorbed dose." *J. Appl. Cryst.* **37**, 513-522.

J. Navaza (1994). "AMoRe: an Automated Package for Molecular replacement." *Acta Cryst. A***50**: 157-163.

C. Nave & E. Garman (2005). "Towards an understanding of radiation damage in cryocooled macromolecular crystals." *J. Synchrotron Rad.*, **12**, 257-260.

J. Ohana, L. Jacquamet, J. Joly, A. Bertoni, P. Taunier, L. Michel, P. Charrault, M. Pirocchi, P. Carpentier, F. Borel, R. Kahn & J.-L. Ferrer (2004). "CATS: a Cryogenic Automated Transfer System installed on the beamline FIP at ESRF" *J. Appl. Cryst.* **37**, 72-77.

Z. Otwinowski & W. Minor (1997). "Processing of X-ray diffraction data collected in oscillation mode." *Methods Enzymol.* **276**, 307-326.

A. Patterson (1935). *Zeitschrift für Kristallographie* **90**, 517.

M. Purdy, P. Ge, J. Chen, P. Selvin & M. Wiener (2002). "Thiol-reactive lanthanide chelates for phasing protein X-ray diffraction data." *Acta Cryst. D***58**, 1111-1117.

U. Ramagopal, M. Dauter & Z. Dauter (2003a). "SAD manganese in two crystal forms of glucose isomerase." *Acta Cryst. D***59**, 868-875.

U. Ramagopal, M. Dauter & Z. Dauter (2003b). "Phasing on anomalous signal of sulfurs: what is the limit ?" *Acta Cryst. D***59**, 1020-1027.

R. Ravelli, H. Schröder Leiros, B. Pan, M. Caffre & S. McSweeney (2003). "Specific Radiation Damage Can Be Used to Solve Macromolecular Crystal Structures." *Structure*, **11**, 217-224.

- R. Ravelli & S. McSweeney (2000). "The fingerprint that X-rays can leave on structures." *Structure*, 8, 315-328.
- P. Retailleau, N. Colloc'h, D. Vivares, F. Bonnete, B. Castro, M. El Hajji & T. Prange (2005). "Urate Oxidase from *Aspergillus Flavus*: New Crystal-Packing Contacts in Relation to the Content of the Active Site." *Acta Cryst.* D**61**, 218-229.
- G. Rieck (1985). "Tables Relating to the production, wavelengths and intensities of x-rays." in *International Tables for X-ray Crystallography, III, X-rays and their interaction with crystals*. Dordrecht (Holland): D. Reidel Publishing Company for International Union of Crystallography, 59-72.
- M. Rossmann (1972). Editor. *The Molecular Replacement Method*. New York: Gordon and Breach.
- R. Sanishvili, A. Joachimiak, A. Edwards, A. Savchenko & T. Skarina "Putative Ribosomal Protein" en prep.
- D. Sayre (1952) "The squaring method: a new method for phase determination." *Acta Cryst.* **5**: 60-65.
- M. Schiltz, P. Dumas, E. Ennifar, C. Flensburg, W. Paciorek, C. Vonrhein & G. Bricogne (2004). "Phasing in the presence of severe site-specific radiation damage through dose-dependent modelling of heavy atoms." *Acta Cryst.* D**60**, 1024-1031.
- M. Schiltz, R. Fourme & T. Prangé (2003). "Use of noble gases xenon and krypton as heavy atoms in protein structure determination." *Methods Enzymol.* **374**, 83-119.
- M. Schiltz, Å. Kvik, O. Svensson, W. Shepard, E. de LaFortelle, T. Prangé, R. Kahn & R. Fourme (1997). "Protein Crystallography at Ultra-short Wavelengths : Feasibility Study of Anomalous Dispersion Experiments at the Xenon K-Edge." *J. Synchrotron Rad.* **4**, 287-297.
- T. Schneider & G. Sheldrick (2002). "Substructure solution with SHELXD." *Acta Cryst.* D**58**, 1772-1779.
- A. Schuettkopf & D. van Aalten (2004). "PRODRG - a tool for high-throughput crystallography of protein-ligand complexes." *Acta Cryst.* D**60**, 1355-1363.
- G. Sheldrick (1998). In *Direct Methods for Solving Macromolecular Structures*. Edited by Fortier S. Dordrecht, The Netherlands: Kluwer Academic Publishers; 401-411.
- T. Terwilliger (2002). "Automated main-chain model-building by template-matching and iterative fragment extension." *Acta Cryst.* D**59**, 34-44.
- T. Terwilliger & J. Berendzen. (1999). "Automated MAD and MIR structure solution." *Acta Cryst.* D**55**, 849-861.
- I. Usón & G. Sheldrick (1999). "Advances in direct methods for protein crystallography." *Current Opinion in Structural Biology*, **9**, 643-648.



M. Vaney, S. Maignan, M. RiesKautt & A. Ducruix, A. (1996). "High-resolution structure (1.33 angstrom) of a HEW lysozyme tetragonal crystal grown in the APCF apparatus. Data and structural comparison with a crystal grown under microgravity from SpaceHab-01 mission." *Acta Cryst.* **D52**, 505-517.

J. Victoreen (1949). "The Calculation of x-ray Mass Absorption Coefficients." *Journal of Applied Physics* **20**, 1141-1147.

D. Vivarès & F. Bonneté (2002). "X-ray scattering studies of *Aspergillus flavus* urate oxidase: towards a better understanding of PEG effects on the crystallization of large Proteins." *Acta Cryst.* **D58** 472-479.

C. Weeks & M. Miller (1999). "The design and implementation of SnB v2.0." *J. Appl Crystallogr* **32**, 120-124.

M. Weik, R. Ravelli, G. Kryger, S. McSweeney, M. Raves, M. Harel, P. Gros, I. Silman, J. Kroon & J. Sussman (2000). "Specific chemical and structural damage to proteins produced by synchrotron radiation." *Proc. Natl. Acad. Sci.* **97**, 623-628.

M. Weiss, G. Mander, R. Hedderich, K. Diederichs, U. Ermler & E. Warkentin (2004). "Determination of a novel structure by a combination of long-wavelength sulfur phasing and radiation-damage-induced phasing." *Acta Cryst.* **D60**, 686-695.