This Bijvoet difference, however, is still small and highly accurate data collection is essential. One of the experimental difficulties using longer wavelength is the increased absorption. Therefore we have developed a crystal mounting technique to eliminate absorption by the frozen cryo-buffer around protein crystal (Kitago et al., 2005), and the practical applicability of this mounting method was examined using several novel proteins at CrKα radiation of 2.29 Å (Watanabe, 2006). In order to utilize this mounting method at the synchrotron beamlines, we made this mounting tool compatible to the standard Hampton CrystalCap. With a special magnet base for this cap, it becomes possible to mount and remount frozen crystals as the standard CrystalCap. We have tested its applicability by sending several frozen crystals to the beamline BL13B1 at NSRRC, Taiwan. This mounting method is also very useful at synchrotron beamlines to mount tiny crystals that are difficult to center because of the lens-shaped frozen buffer in the cryoloop. In this development, we also use a loop made of a polyimide film microfabricated by photolithography.

[1] Kitago, Y., Watanabe, N. and Tanaka, I. (2005), *Acta Cryst*., **D61**(8), 1013-1021.
[2] Watanabe, N. (2006), *Acta Cryst*., **D62**(8), 891-896.

Keywords: SAD, longer wavelength SAD, crystal mounting method

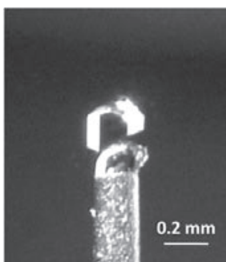
P04.10.262

Acta Cryst. (2008). A**64**, C313

Application of a novel mounting tool using adhesive for protein crystals

Tomoya Kitatani[1,5], Tomokazu Hasegawa[2], Sugiyama Shigeru[1,5], Hamada Kensaku[2], Matsumura Hiroyoshi[1,4,5], Takano Kazufumi[1,4,5], Adachi Hiroaki[1,4,5], Murakami Satoshi[3,4,5], Inoue Tsuyoshi[1,4,5], Mori Yusuke[1,4,5]
[1]Osaka University, Department of Electrical Engineering, Graduate School of Engineering, 2-1 Yamadaoka, Suita, Osaka, 565-0871, Japan, [2]PharmAxess, Inc., Osaka 567-0085, Japan, [3]The Institute of Scientific and Industrial Research, Osaka University, Ibaraki, Osaka 567-0047, Japan, [4]SOSHO Inc., Osaka 541-0053, Japan, [5]CREST JST, Suita, Osaka 565-0871, Japan, E-mail：kitatani@chem.eng.osaka-u.ac.jp

We have developed a novel mounting tool for protein crystals that directly captures a crystal from a drop with an adhesive (Kitatani *et al*., 2008). This tool has a loop-free structure and can reduce the amount of solution around the captured crystal in comparison with conventional loop-based mount tools. The extra solution around captured crystal not only degrades the quality of data-collection statistics but also disturbs the initial phase determination by the sulfur single-wavelength anomalous diffraction (SAD) method. The effect of reducing the solution in this tool is helpful in the sulfur SAD method. We have evaluated the effects of our tool on a quality and crystallographic data in the sulfur SAD method using CoK_α radiation.



Reference

[1] Kitatani T., Sugiyama S., Matsumura H., Adachi H., H. Yoshikawa Y., Maki S., Murakami S., Inoue T., Mori Y. and Takano K. (2008) *Appl. Phys. Express*, 1, 037002.

Keywords: biomaterials and biodevices, protein crystals, SAD

P04.10.263

Acta Cryst. (2008). A**64**, C313

Structural transitions and the evolution of protein crystal disorder during slow cooling

Matt A Warkentin[1], Robert E Thorne[1,2]
[1]Cornell University, 219 Clark Hall, Ithaca, NY, 14853, USA, [2]Mitegen LLC, Ithaca, NY, 14850, USA, E-mail：maw64@cornell.edu

We report measurements of the lattice perfection and unit cell volume of Lysozyme, Thaumatin, and Trypsin crystals as function of temperature during slow cooling (200 K/hour) from 300 K to 100 K. We demonstrate that these crystals can be slow cooled without formation of internal crystalline ice, even when no penetrating cryoprotectants are used. Most degradation of diffraction properties occurs between 240 and 180 K. In Thaumatin crystals, slow cooling produces low temperature diffraction properties that are as good as or better than those obtained with flash cooling or with cooling under pressure. In Lysozyme and Trypsin crystals, crystal order abruptly and dramatically degrades near 210 K. This dramatic degradation is associated with an apparent first-order phase transition signaled by an abrupt decrease in unit cell volume. We examine two possible origins for this transition: cold denaturation of the protein, and a transformation between low density and high density liquid water within the nanometer-sized channels of the crystal.

Keywords: cryocooled crystallography, structural phase transitions, biological macromolecular crystallography

P04.10.264

Acta Cryst. (2008). A**64**, C313

The temperature dependence of radiation damage to macromolecular crystals

Robert E Thorne[1,2], Matthew A Warkentin[1]
[1]Cornell University, Physics Department, 219 Clark Hall, Ithaca, NY, 14853, USA, [2]Mitegen LLC, Ithaca, NY, 14850, USA, E-mail：ret6@cornell.edu

We have studied how the rate of global radiation damage to Lysozyme, Thaumatin, and Trypsin crystals varies with temperature. The radiation sensitivity of these proteins decreases by a factor of 30 to 50 between T=300 K and 100 K. Most of the decrease occurs between 220 K and 180 K, and we attribute this to a glass transition in the protein + solvent system. Addition of 40% glycerol raises the temperature for the drop in radiation sensitivity by ~10 K, consistent with the measured shift in the glass transition.

Keywords: radiation damage, biological macromolecular crystallography, glass transition

P04.10.265

Acta Cryst. (2008). A**64**, C313-314

Manipulating protein microcrystal with optical tweezers based on lensed fiber probes

Takaaki Hikima, Tetsuya Shimizu, Masaki Yamamoto
RIKEN SPring-8 Center, 1-1-1 Kouto, Sayo-cho, Sayo-gun, Hyogo, 6795148, Japan, E-mail：hikima@spring8.or.jp

In many synchrotron facilities, X-ray microbeam will be utilized for protein crystallography. It will be possible to collect diffraction data from a protein microcrystal with the size in the range from 1