MicroMounts: Crystal Handling and Mounting in the High-Throughput Era

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Thanks to:
NASA, NIH
Mounting Crystals for Flash Cooling and Low-Temperature X-Ray Data Collection

Present Technology: Nylon Loops

1. Difficult to handle crystals smaller than 50µm
2. Irreproducible hole size and shape
3. Undesirably flexible
4. Difficult to retrieve and manipulate crystals
4. Irreproducible crystal positioning; difficult automated alignment
5. Excess liquid complicates cryoprotection, scatters X-rays
Our Answer: MicroMounts

- Consist of a microfabricated polyimide film attached to a round solid stainless steel post
Oxide-coated Silicon wafer with microfabricated polyimide film
Microfabricated thin film of polyimide:
- low Z constituents, low density ⇒ less background scatter than nylon

Film curvature:
- provides excellent stiffness even with very thin (5 micron) films, and a convenient scoop-like action in retrieving and handling crystals

Wicking hole
- allows easy removal of excess liquid

Alignment pattern:
- Allows easy manual and automated alignment
Absorption versus resolution:
25 µm nylon versus 10 µm polyimide
Complete data set to 3.1 Å resolution obtained using a single 5 x 7 micron crystal with a 104 Å unit cell
Advantages over CryoLoops:

- Easy handling of crystals as small as 5 µm.
- Rigid yet thin construction ⇒ reduced bending in cryostreams and viscous solutions
- Convenient, scoop-like action ⇒ reduced chance of crystal loss or damage.
- Crystal hole size and shape precisely determined ⇒ easy mounting and alignment
- Easy removal of excess liquid ⇒ easier cryoprotection, faster freezing
Advantages over CryoLoops:

• Reduced background scatter
  ⇒ higher achievable resolution

• Solid pins
  ⇒ easily bent to place crystal in desired orientation
  ⇒ no wicking of liquid inside the pin
  ⇒ can be cut to any desired length.
Advantages over CryoLoops:

• Easy automated alignment. Beam position and size can be determined without ever optically observing the crystal.
Recent Developments:

MicroMesh™ Holders for Rapid Screening:
A New Approach to Data Collection at Room Temperature

Why collect data at room temperature?

• Some macromolecular crystals not easily frozen

• Diagnose source of poor low-temperature diffraction properties

• Potentially higher resolution and more information (but requires big crystals)
Replace glass with polyethylene (PET) tubing
Reduced diffuse scatter:

- 10 µm quartz
- 50 µm PET
- 25 µm PET
- 12.5 µm PET
Comparison of integrated diffuse scatter:

![Bar graph comparing normalized integrated intensity for different materials. The materials are 10 micron Quartz, 50 micron PET, 25 micron PET, and 12.5 micron PET. The graph shows that 10 micron Quartz has the highest normalized integrated intensity, followed by 50 micron PET, then 25 micron PET, and finally 12.5 micron PET.]
Evaporation through PET tubing:

- 50 µm PET
- 25 µm PET
- 12.5 µm PET
- Double 25 µm PET
- (50µm+25 µm) PET
- Painted 25 µm PET
- Kapton

**Evaporation rate (mg/day)**

**Material and thickness**
As grown

After cryoprotectant soak

After flash cooling
Thanks to:

**MicroMounts:**
Adam Bartnik, Zach Stum, Jan Kmetko, Kevin O’Neill, Richard Gillilan, Andy Stewart

**Room T Mounts:**
Eugene Kalinin, Emil Lobkovsky

**Good Advice:**
Steve Ealick, Tom Ellenberger