Jena Bioscience **Technical Note** MiTeGen

Using MicroMounts™ and MicroLoops™ for Macromolecular Crystallography

MicroMounts™ and MicroLoops™ are used in much the same way as nylon loop mounts, although there are some important differences. The nonmagnetic stainless steel pins are compatible with existing bases and mounting hardware. Our suggested procedures follow. If you find better or novel ways to use them, please tell us!
If you're using the smaller sizes, be sure to read our tips on Handling and Mounting Small Crystals.

- Select a mount with a sample aperture size **slightly smaller** than the crystal size. Unlike with loops, *you don't want to trap the crystal inside a liquid meniscus spanning the aperture*. You want to have it resting on the edges of the aperture, to minimize excess liquid and background X-ray scatter.

- Cut the pin (if necessary) to a desired length using our pin cutters or other cutters designed for spring-temper steel. The center-to-center distance between black lines on the pin is 2 mm. Use the millimeter scale on the top of the box to help cut it to the desired length, lining up the end of the pin with the right end of the scale.

- Insert the pin into a standard goniometer base using a small amount of, e.g., Dow-Corning #976V high vacuum grease or Duco Cement to hold it in place or use reusable goniometer bases instead. For easier handling, attach the base to a magnetic rod. The pin can also be inserted into a 0.7 mm mechanical pencil.

- Slowly and carefully insert the gold-colored tip into the crystal-containing drop, trying to minimize fluid motion. Slide the sample aperture under your crystal, and then carefully remove the crystal + mount from the drop, keeping the crystal centered over the sample aperture.

- To retrieve crystals that have settled to the bottom of a crystallization plate, try pressing downward to bend the tip of the mount so that it slides flat along the bottom.

- If there is excess liquid, slowly and carefully insert a size 15 paper wick into the large opening that connects to the sample hole. If you have lots of liquid and you insert the wick too quickly, the crystal will flow with the liquid to the wick. For viscous liquids you may need to "mop up" around the crystal with the wick.

- You should find that it is no longer necessary to use Paratone or other heavy oils to replace surrounding liquid that otherwise would form hexagonal ice rings. Mounted correctly, **you should have very little liquid around your crystal** - far less than when using nylon loops. The flash cooling rate will be much faster than in a loop and the cryoprotectant concentrations required to prevent icing much smaller. Your penetrating cryoprotectant concentration should often be sufficient to prevent crystallization of external liquid.

- In general, it's bad to have a lot of any liquid - water or oil - around your crystal during flash cooling: when the liquid freezes, it will tend to crush your crystal. If you have just a thin layer of liquid around the crystal, it will shatter (like the shell of an egg) and so do little damage.

- Flash cool the crystal by your favorite method. We recommend plunge cooling in liquid nitrogen, in liquid nitrogen that has been vacuum-pumped to near its freezing temperature (to reduce boiling), or (somewhat better but much more dangerous) in liquid propane.

Please contact xtals@jenabioscience.com with comments or suggestions.