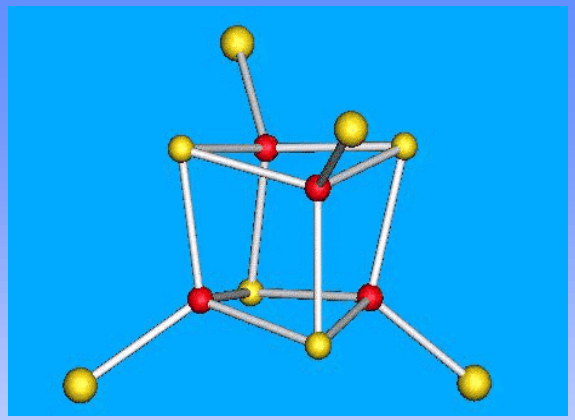


X-ray Crystallography: Lecture 4:

Crystals Growing, Screening and Mounting

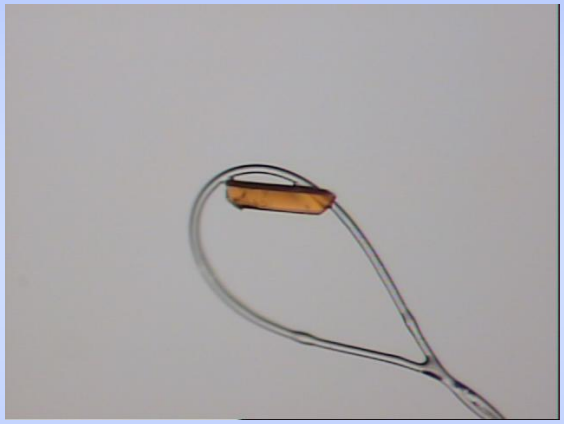
Prof. S. K. Gupta

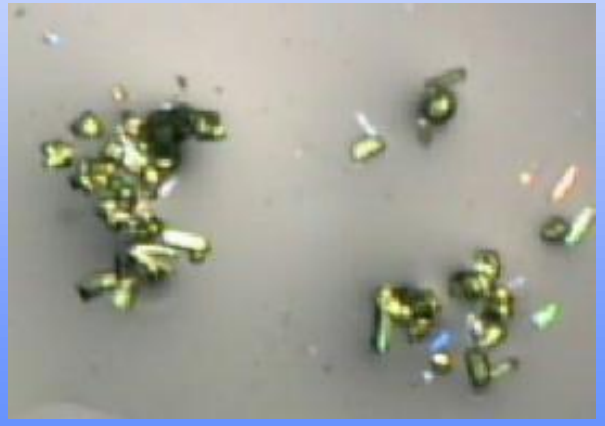
Why are you interested

- You are research chemists
- Most will be involved in chemical synthesis (organic, inorganic, organometallic, biochemical)
- Determination of structure a key goal

What do I need to supply to the Crystallographer?

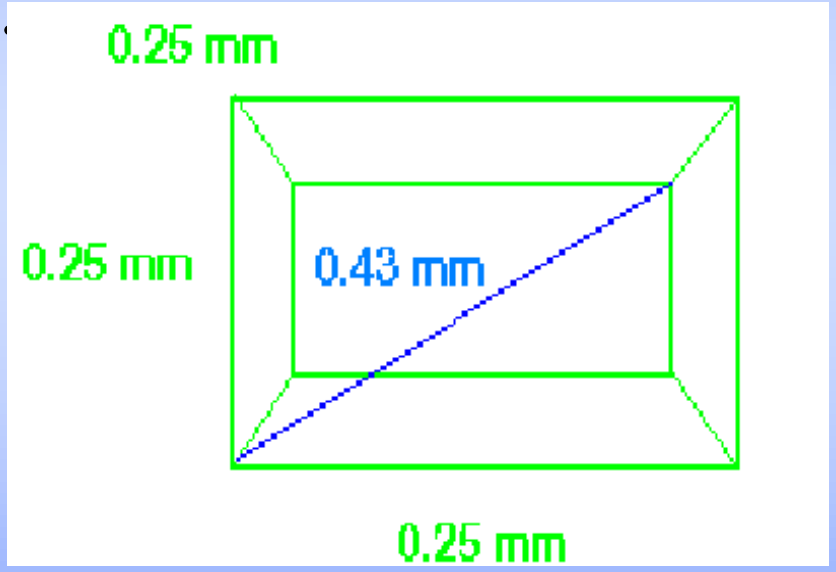
- Single Crystals
- Bring what you can grow
- Chemical Formula
- Compound Name
- Solvents/conditions used
- If not single--- Discuss recrystallization





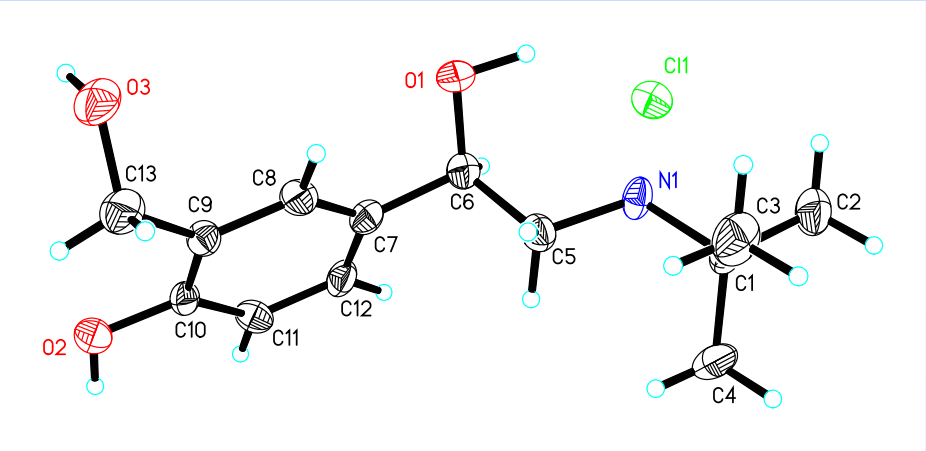
Crystal Size

- Size should be 0.25 x 0.25 x 0.25 mm, perfect.
- Gives 0.43 mm diagonal.
- Smaller crystals are very possible!
- Larger crystals can be cut!



Where Do I Start?

- Simple Recrystallization.
- During purification did you create crystalline material?
- Are these crystals Big enough?



These crystals were 0.05 x 0.025 x0.002 mm

How much Material Do You Need?

- Depends on the vessel you are going to use to grow the crystals.
- Depend on solubility of sample in the solvent.
- NMR sample generally a good concentration level.

How much Material is in a Single Crystal?

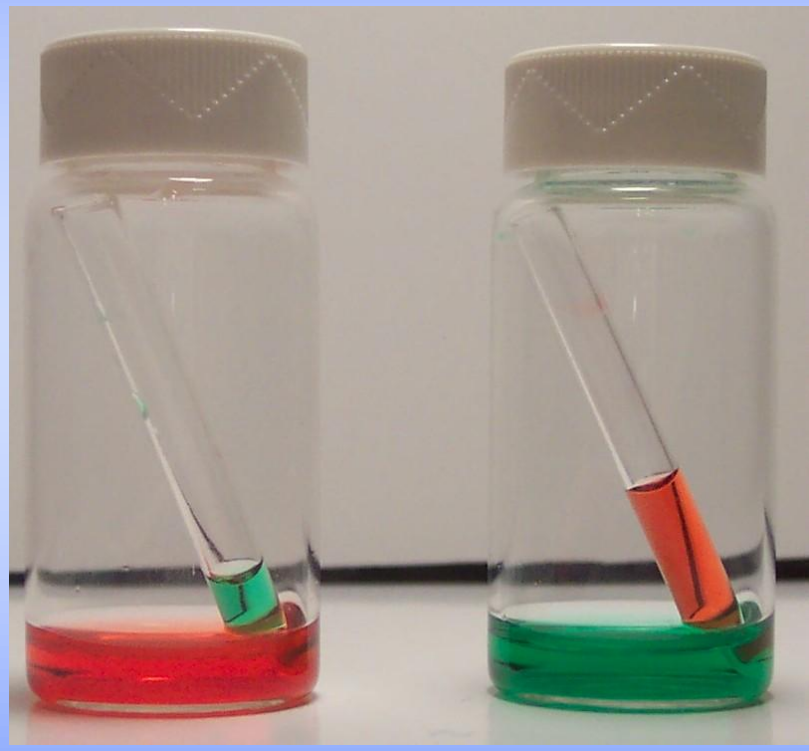
- If the crystal for x-ray diffraction is to be 0.3 x 0.3 x0.3 mm, volume = 0.027 mm^3
- Typical unit cell is 12 x 12 x12 Å; volume = 1728 $Å^3$
- Å = 10-10 meters = 10^{-8} cm = 100 pm (picometers)
- Therefore in a typical crystals 1.6 x 10^{16} unit cells
- 1.3 x 10^{17} molecules for 8 molecules per cell.
- MW= 206.2 then only 2.49 x 10^{-7} moles in the cell. 5.1 x 10^{-5} g, 0.051 mg
- Unfortunately more than one crystal grows in the vessel so more material is needed.

What is the Goal

- To create a single crystal which diffracts on the instrument such that an analysis can be accomplished.
- Generally this means trying to get the material to go from solution to a solid very slowly.
- Create an environment that slowly changes over time to cause crystallization.

Vapor Diffusion: What do I grow the Crystals In?

- Clean glassware, most of the time.
- Consider location
- Consider volume needed to grow the crystal.
- Usually clean new vials that fit inside one another work well.

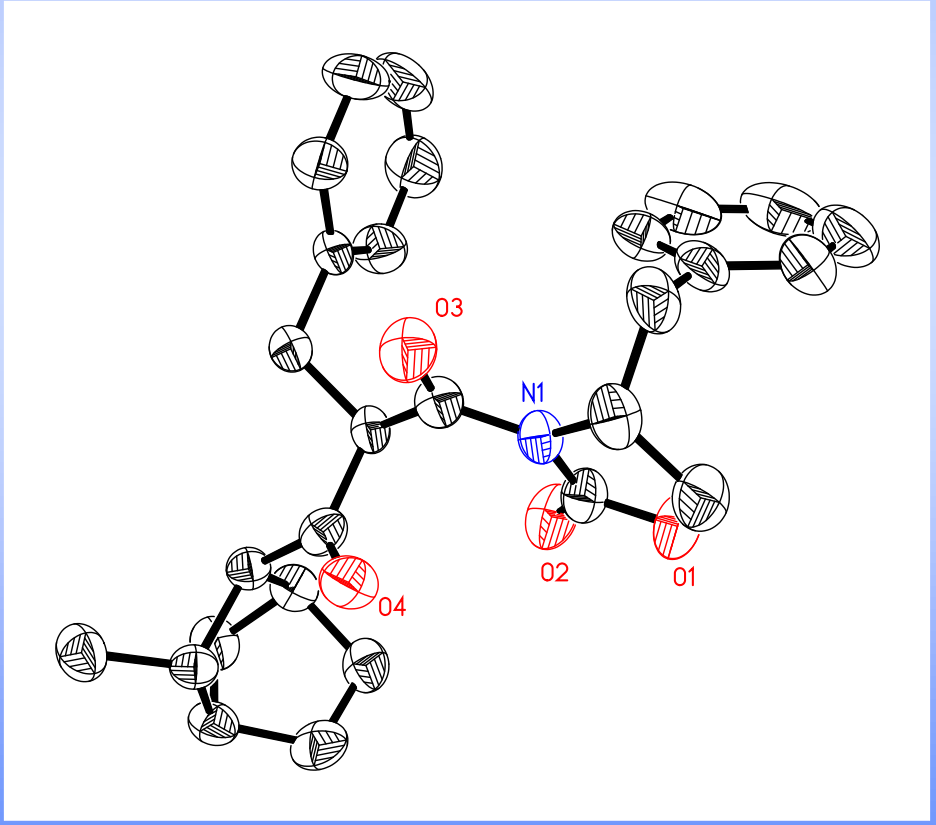


Vapor Diffusion

- Good for milligram amounts.
- Volatile solvents.
- Slowly create a less desirable solvent.
- Need to be aware of vapor pressures of solvents.
- Have available a chart of physical properties of solvents

Example of a Crystallized Compound from Vapor Diffusion

- Used a diffusion chamber with compound in dichloromethane and then hexane in the outside chamber.



Solvent Layering

- Layering must be very careful.
- Place a solvent between the two layers.
- Do not disturb the vessel.
- Set it so you can view it without moving it.

Solvent Choice

- Polar--- polar solvent layered with a non-polar solvent
- Non-polar --Non-polar solvent, evaporation or layer with polar solvent, harder.
- Consider densities, works best with solution as bottom layer

Hydrogen Bonding

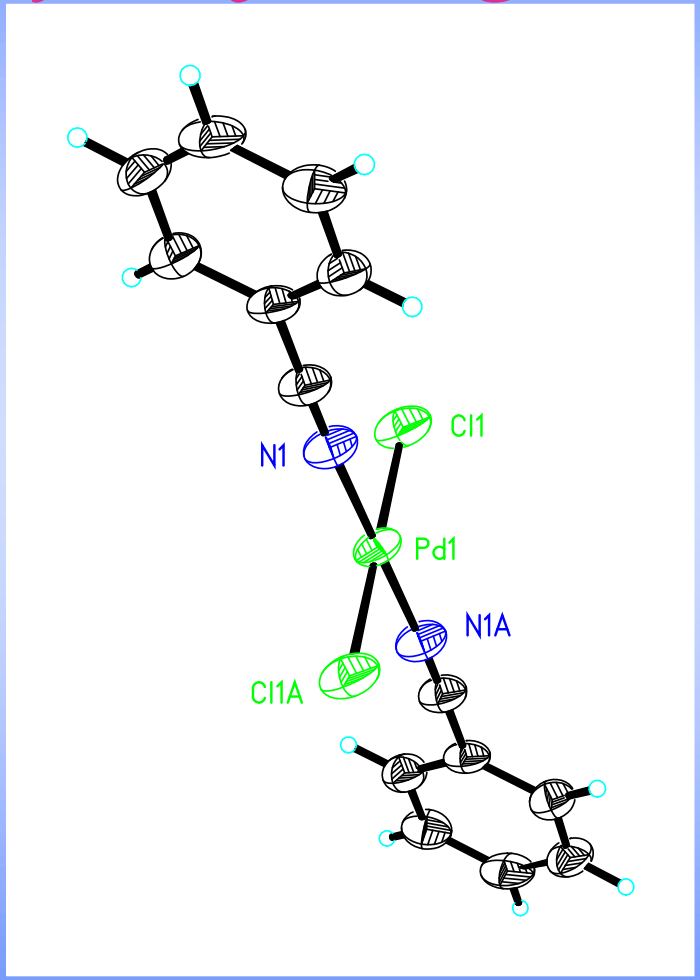
- Hydrogen Bonding is very important in the crystallization process.
- Consider whether hydrogen bonding solvent might help or hinder crystallization.
- Amides generally do better with hydrogen bonding solvents.

Solvents to Use and NOT to Use

- Use Benzene or toluene! Seems to be a magic solvent.
- Aromatic rings seems to help fill holes in lattice as well.
- 2-Butanone works well for organics
- DMF/Ether or CH_3CN/Ether works well for inorganics
- Avoid volatile solvents, CH_2Cl_2, Diethyl Ether as solvent as it evaporates to fast.
- Avoid long alkyl chains, cause disorder.

Example of Layering

- Grown by layering a solution of methylene chloride with pentane.



Reactant Diffusion

- Perform the reaction on a small scale compared to surface area.
- Layer one reactant on top of the other reactant and allow diffusion to control reaction rate and crystal formation.
- Good when product formed is highly insoluble.

Old Stand by: Slow Evaporation

- Filter to remove any particles
- Allow the material to crystallize out as the solvent evaporates.
- Keep the solution clean and covered to avoid dust particles.

Use The NMR Tube

- Often crystals have been received by allowing the solvent to evaporate slowly from the NMR tube (often over a long time – D_6-DMSO).

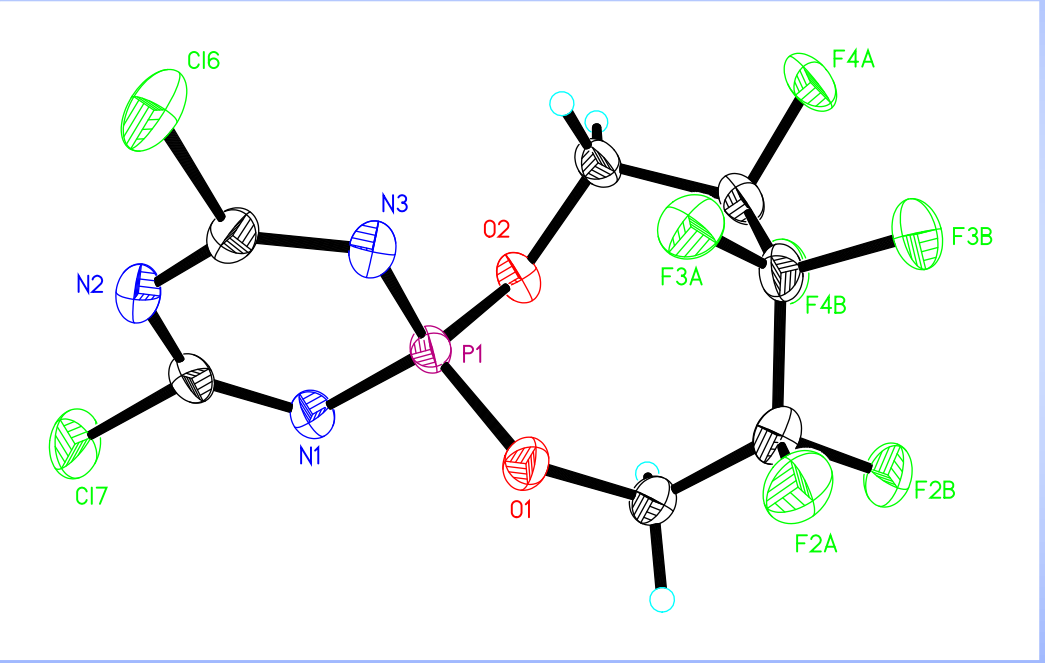
- Remember to keep the tube covered to avoid dust and dirt.

Slow Cooling

- Standard recrystallization technique.
- Must perform this slowly to work well.
- Slow reduction of the temperature is best (programmable hot plate).

Sublimation

- Works extremely well when can be done.
- Must be performed slowly to achieve good size crystals.



Micro Beakers

Micro Beakers (cont'd)

Capacity (ml)	Dimensions (mm)	
0.5	12	7
1.0	12	13 (the best size)
2.5	21	13
5.0	25	19
7.5	25	25

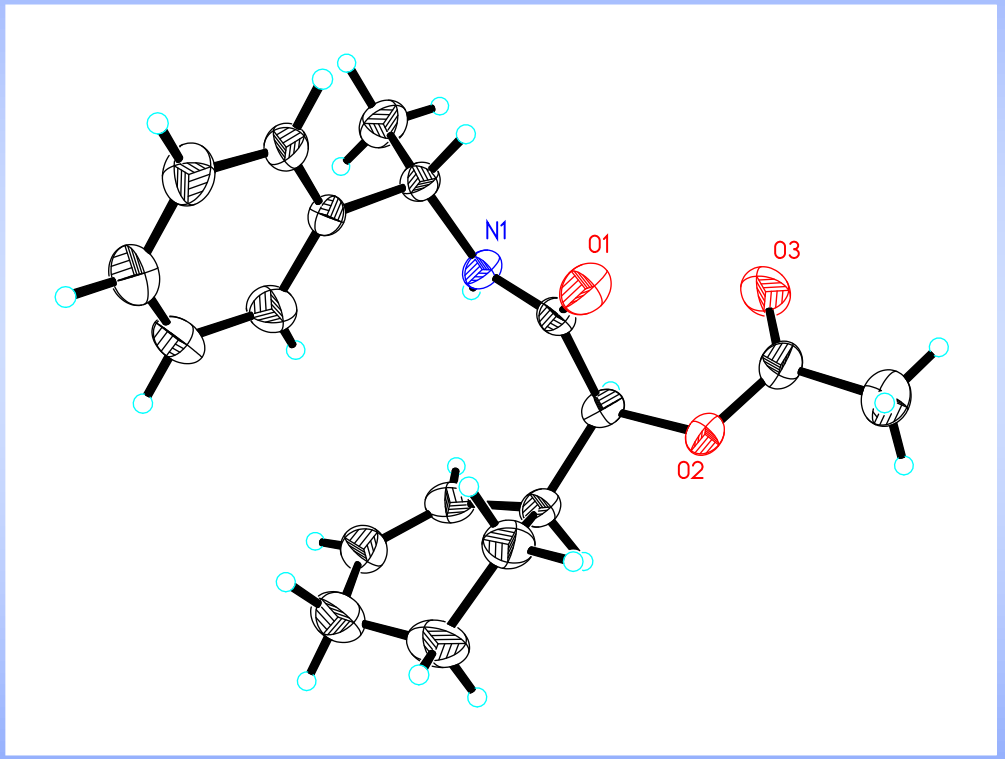
- Excellent for screening as uses very little sample or solvent
- Use microscope cover slip to keep out dust, etc.

Use of derivatives

- Margaret Etter pioneered the use of triphenyl phosphine oxide to obtain crystals in difficult cases
- Forms adducts with hydrogen bonding donor molecules
- Generally forms excellent crystals

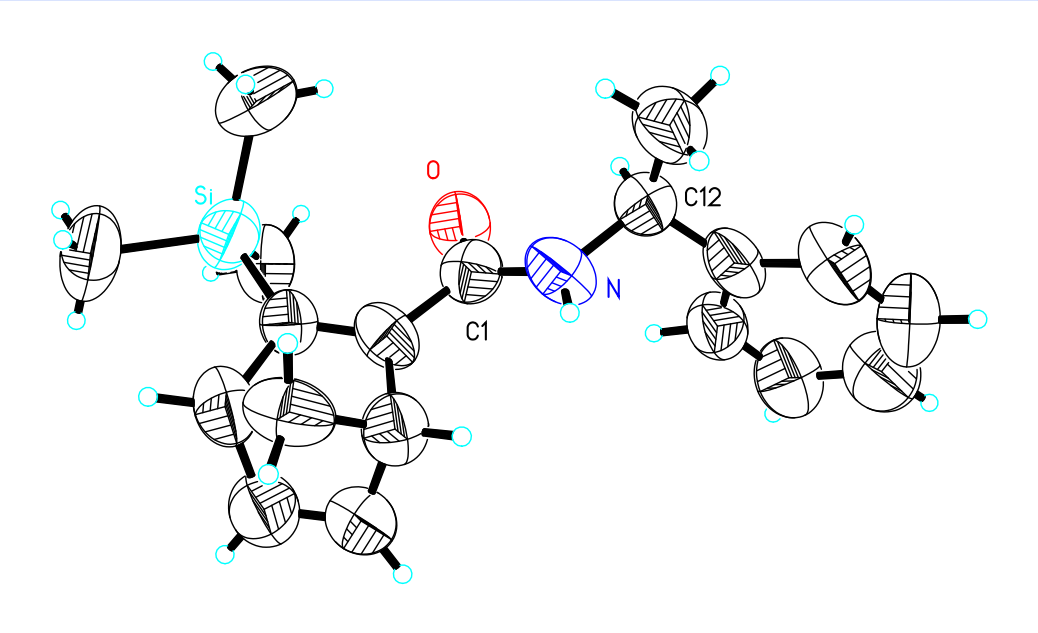
Chiral Compounds

- These tend to be more difficult.
- Try to make derivatives which will improve packing. i.e. phenyl rings.
- Have atoms heavier than carbon.



S-α-methylbenzylamine

- Use with carboxylic acids, could be generated from alcohol or aldehydes.
- Cheap and usually easily crystallized.

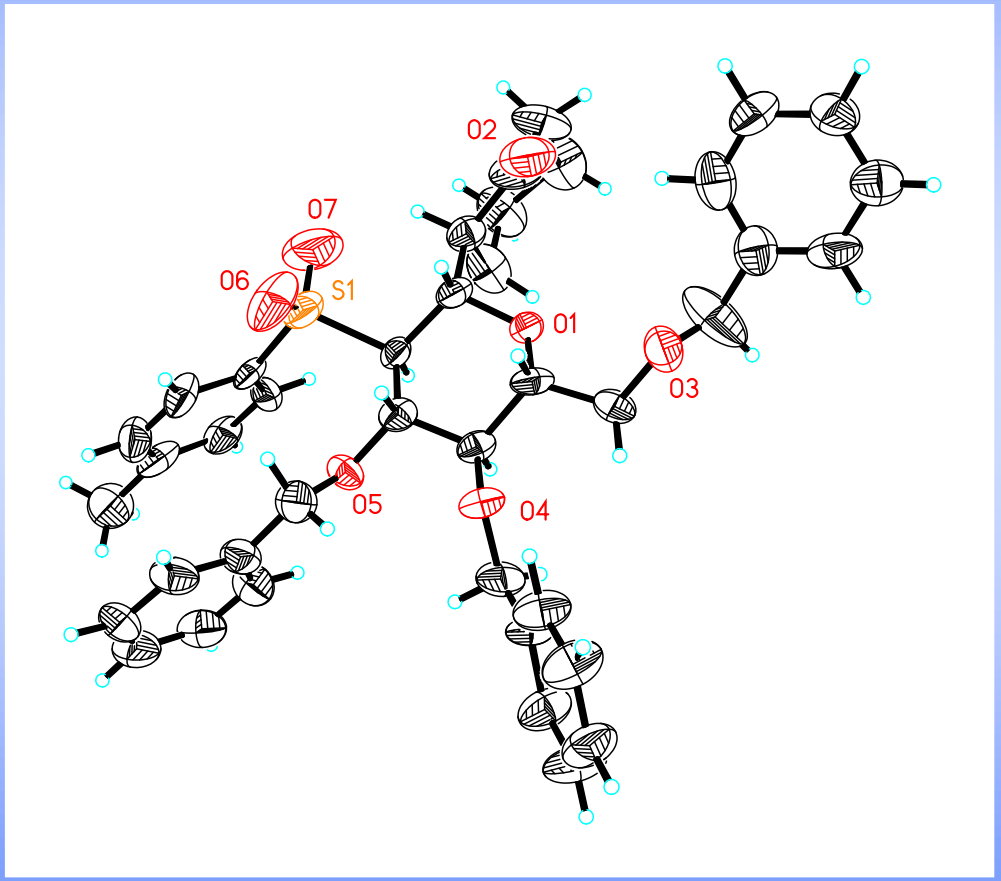


Aldehyde converted to acid then to the amide.

Improve with heavy atom and crystallization

- Have heavy atom present.
- Alcohols and amines make derivatives with

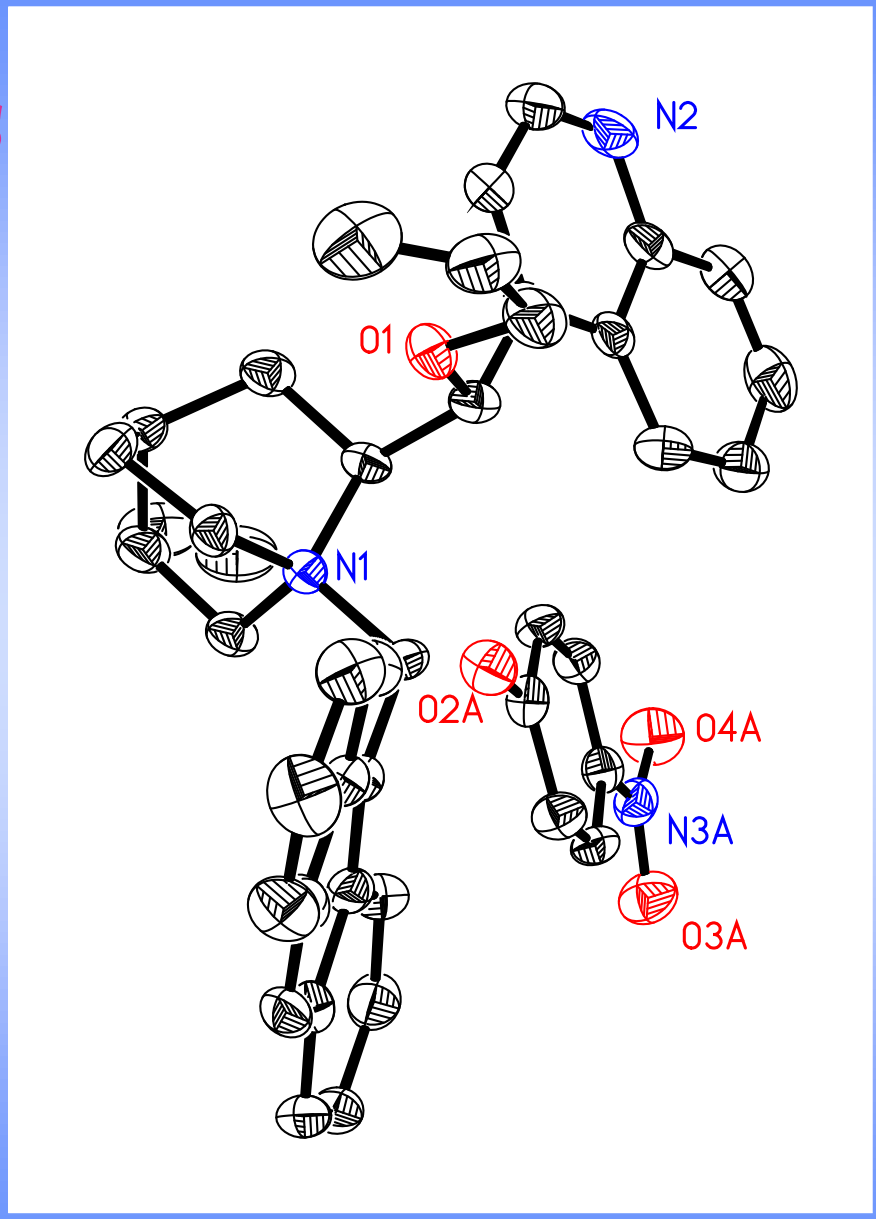
  p-Bromobenzoate
- Include aromatic components in derivative.



Crystal was 0.1 x 0.05 x 0.05 mm, grown benzene layered with hexane.

Counterions

- Change a counterion in the complex.
- Ions of the same size tend to pack well.
- If neutral compound does not crystallize or is a liquid, create an ion.
- Deprotonation or protonation. Good to confirm the identity of the material.



Macro Type Methods applied to small molecules

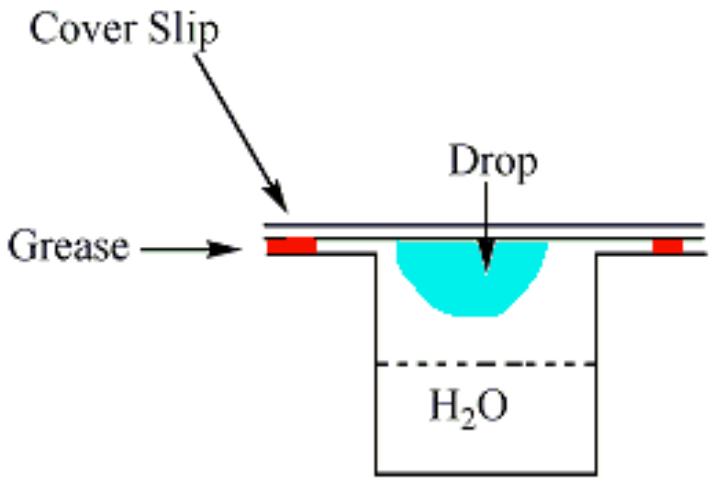
- Hampton Research one of the first companies to address small molecules using macro techniques
  - Problem, organic solvents, not water
- Solution to use alcohols and small quantities of organics
- Success at Howard has been limited

New plates and bridges

- Formulation of new plates and bridges
- Polypropyene
- They have developed some great initial starting solutions. Down load small molecule catalog.

http://www.hamptonresearch.com

Plate Crystallizations, Hanging Drop



Drop Contains

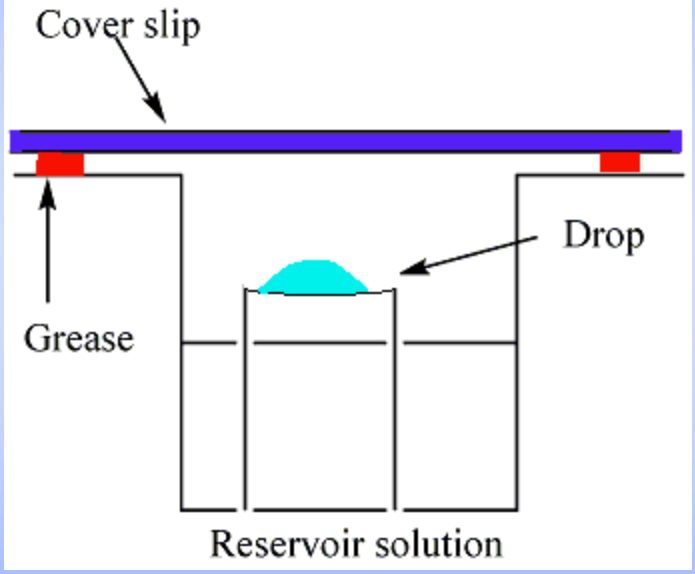
Compound
H_2O
Solvent A

Over time the drop loses Solvent A to the lower reservoir and compound crystallizes

Solvent A = Organic, water soluble solvent, Alcohols, CH_2Cl_2

See Hampton Research catalog or web sit for a very good tutorial on crystal growing by these methods. http://www.hamptonresearch.com

Plate Crystallizations, Sitting Drop

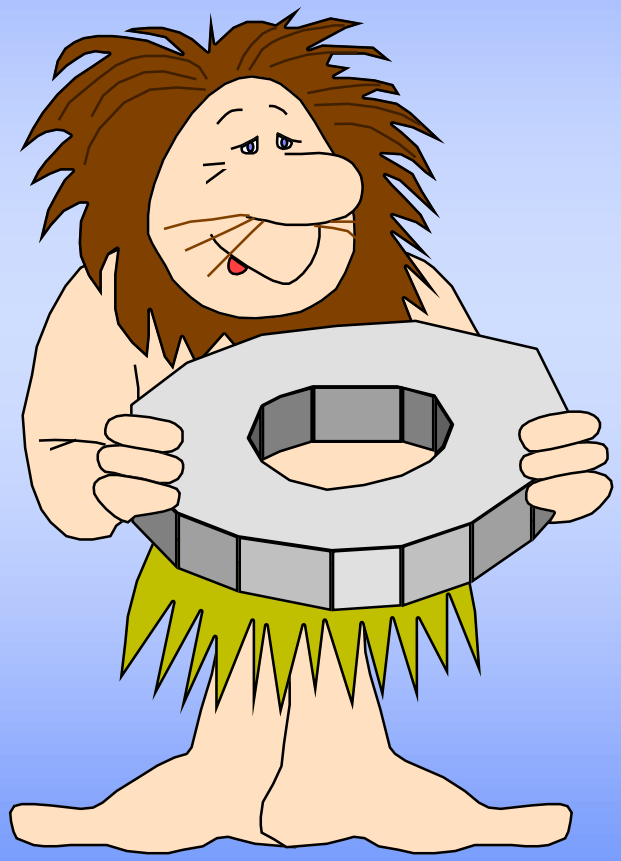


See Hampton Research catalog or web sit for a very good tutorial on crystal growing by these methods. http://www.hamptonresearch.com

Key Factors to Good Crystals.

- Solvent
- Nucleation
- Mechanics
- Time

- Patience, Patience
- Art Form



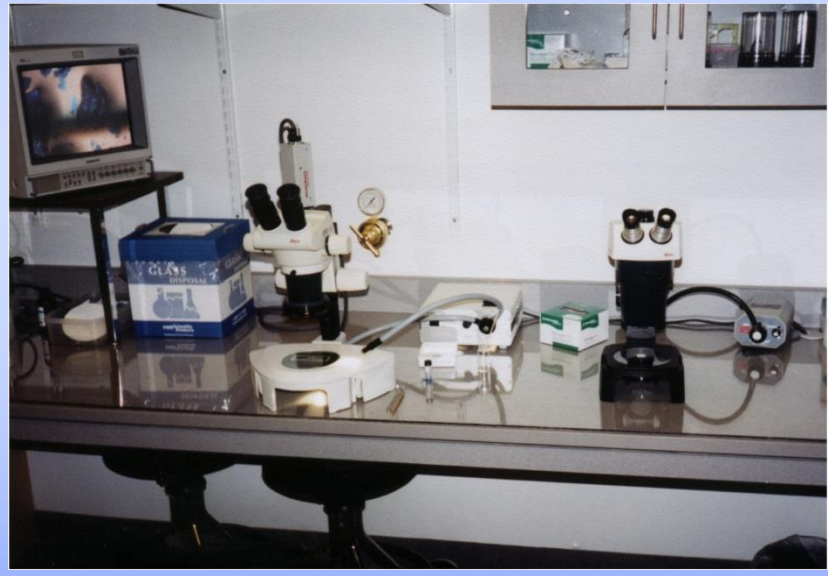
Techniques for Growing Crystals

- For good discussion of key factors in obtaining good crystals.
- Read: "Crystal Growing", Peter G. Jones,

  Chemistry in Britain, *17*(**1981**) 222-225.
- See www site by Paul D. Boyle (http://www.xray.ncsu.edu/GrowXtal.html).

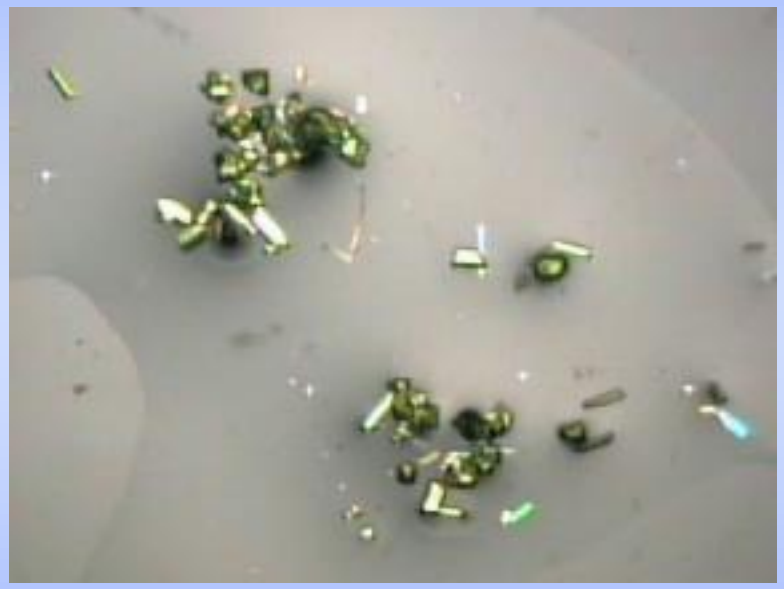
Crystal Evaluation

- Evaluation starts at the microscope. Do they look crystalline and single under cross polarized light?
- Are all the crystals uniform in shape and color -morphology
- Mount and evaluate the crystal on the diffractometer. Requires about 10 - 20 minutes. Less if it does not diffract at all.



What is a Good Crystal?

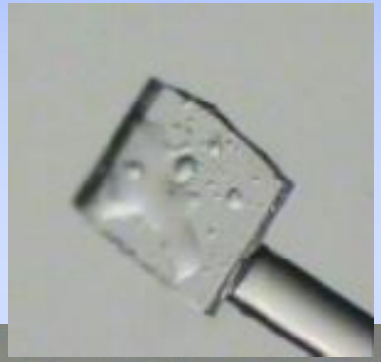
- Well defined crystalline shape often results in good crystals.
- Sparkle
- One that works!!
- Gives good diffraction spots and spot shapes.

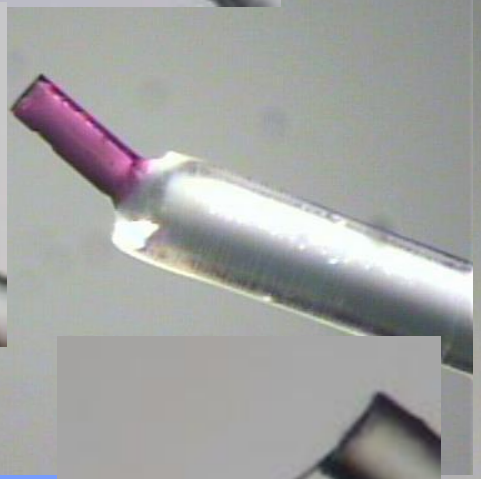
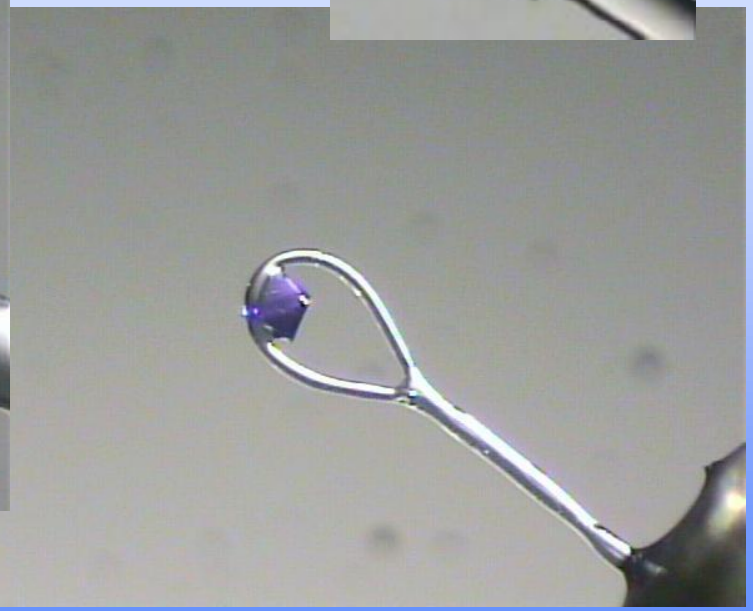
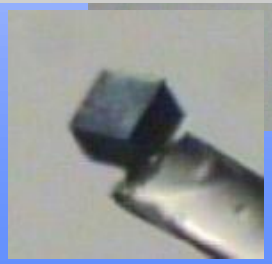




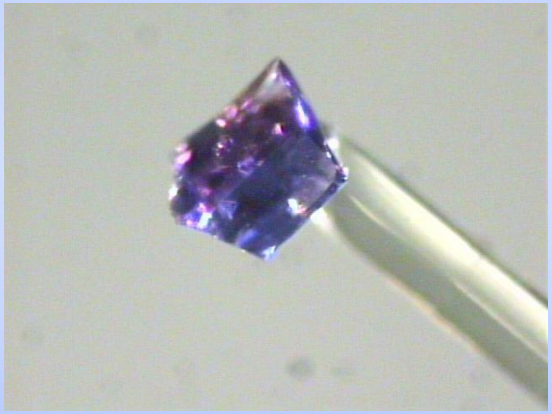




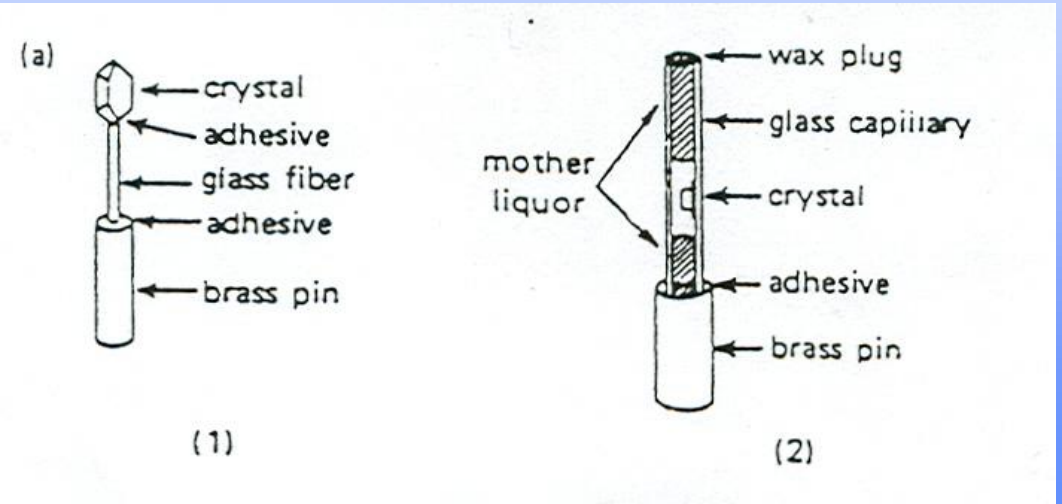

What happens to larger crystals?

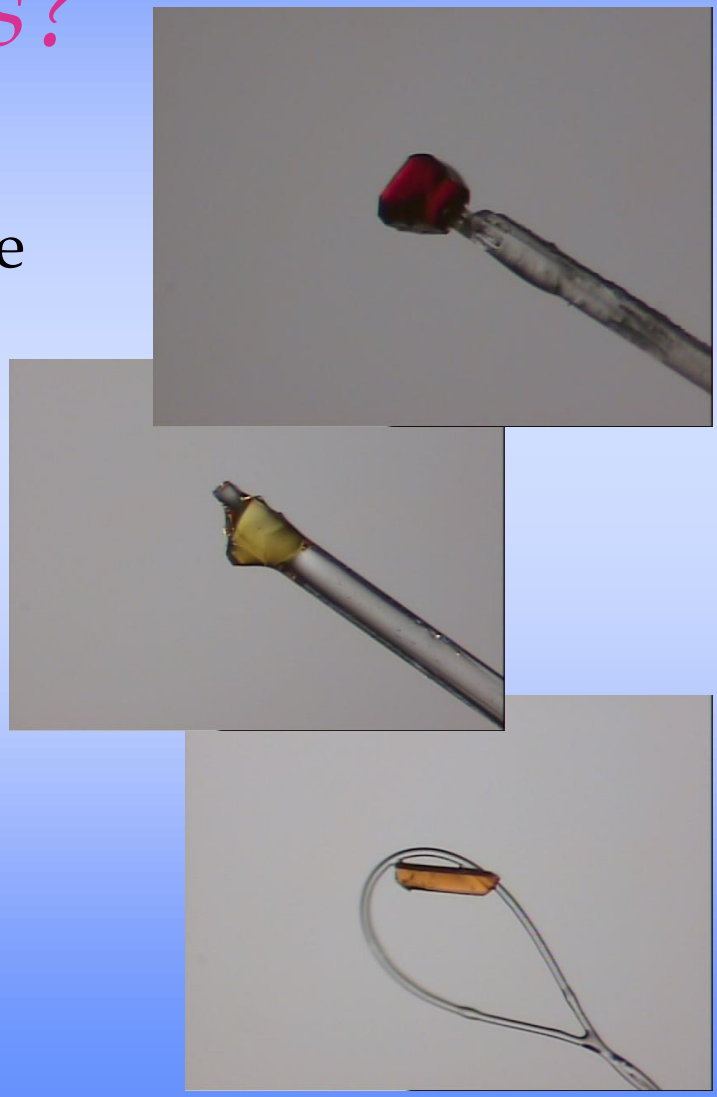


- Cut the crystals to size.
- When cutting do they crumble, these are not likely single.
- Do they start well but become less defined over time, loss of solvent.

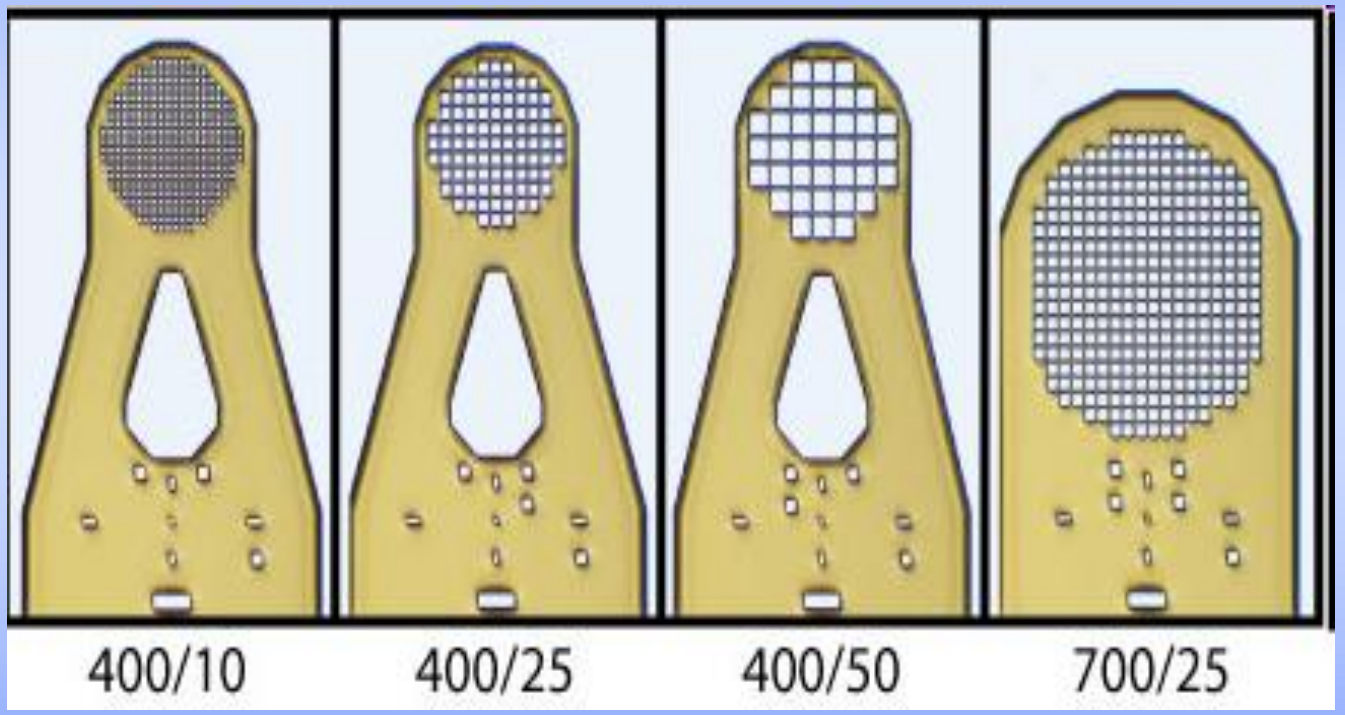
How do you mount the crystals?

- Use 5-min expoxy
  - (Room Temperature)
- Use a small amount of paratone oil or mineral oil. Low Temperature
- Very small crystals, try using a loop.





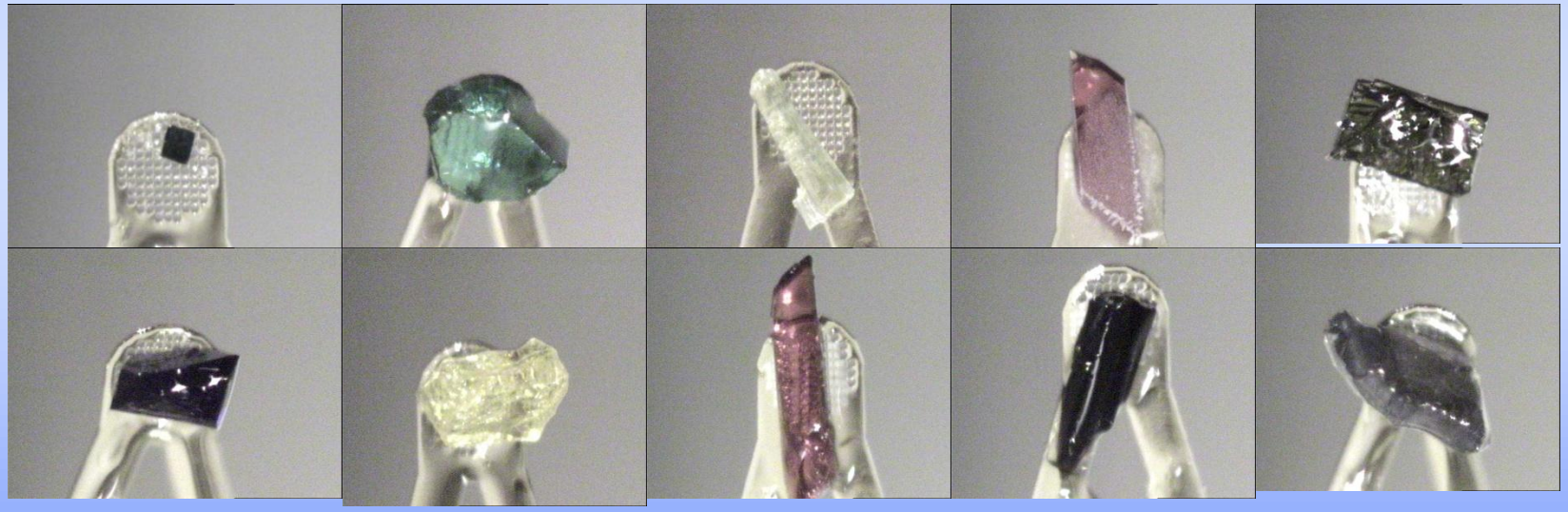
MicroMesh Mounting



MicroMesh cont'd

- MicroMesh Mounts are the tool of choice for microcrystal crystallography and diffraction experiments, but good for any crystal.
- They are excellent for rod/needle shaped crystals, as mesh provides continuous, gentle support for rods and fragile thin plates of all sizes.
- MicroMeshes produce the smallest background scatter of any commercial mount.
- Their sieve-like action allows easy retrieval of crystals from oil (fishing).

Examples, from small to large



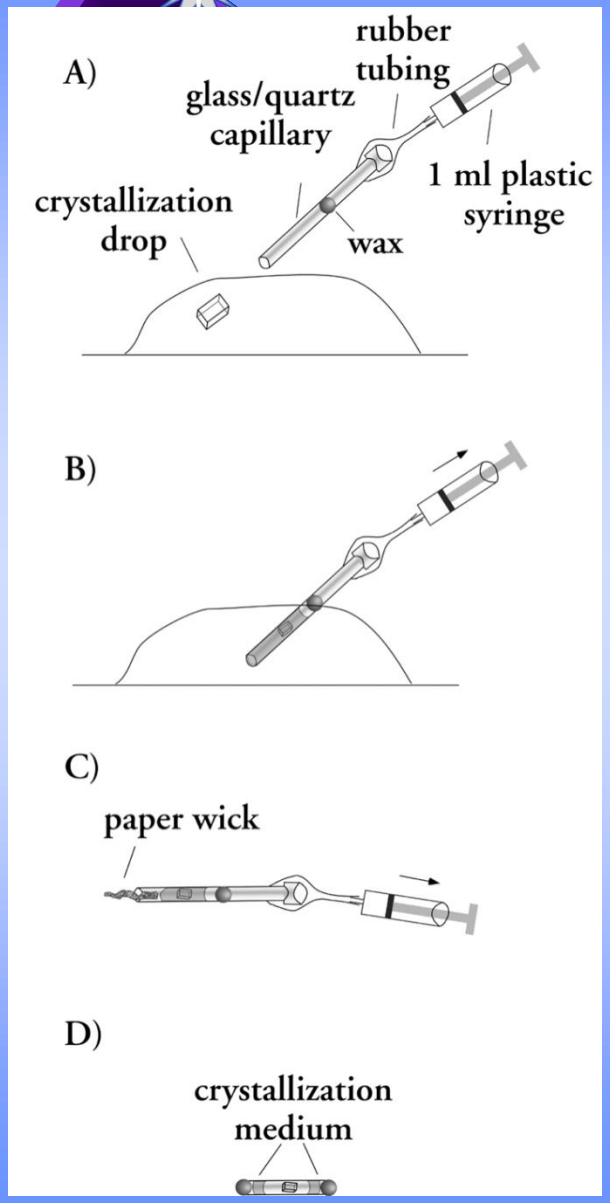
Getting Crystal on Fiber or Loop

All Crystallographers have their own method to accomplish this task. Some will work for you and some may not work as well. These two methods work for most students at Howard, running low temperature data collections.

Slide method Place the crystals in a small amount of Paratone oil. Slide the crystal out of the oil until the crystal prefers to stay on the fiber. This works well for large crystals mounted on fibers.

Pick up Try and put enough oil on the fiber that the crystal comes with the oil. This method means you should try to remove excess oil before placing on diffractometer. Good for loops. Slide method also works for loops with small crystals.

Mounting Crystals In Capillaries



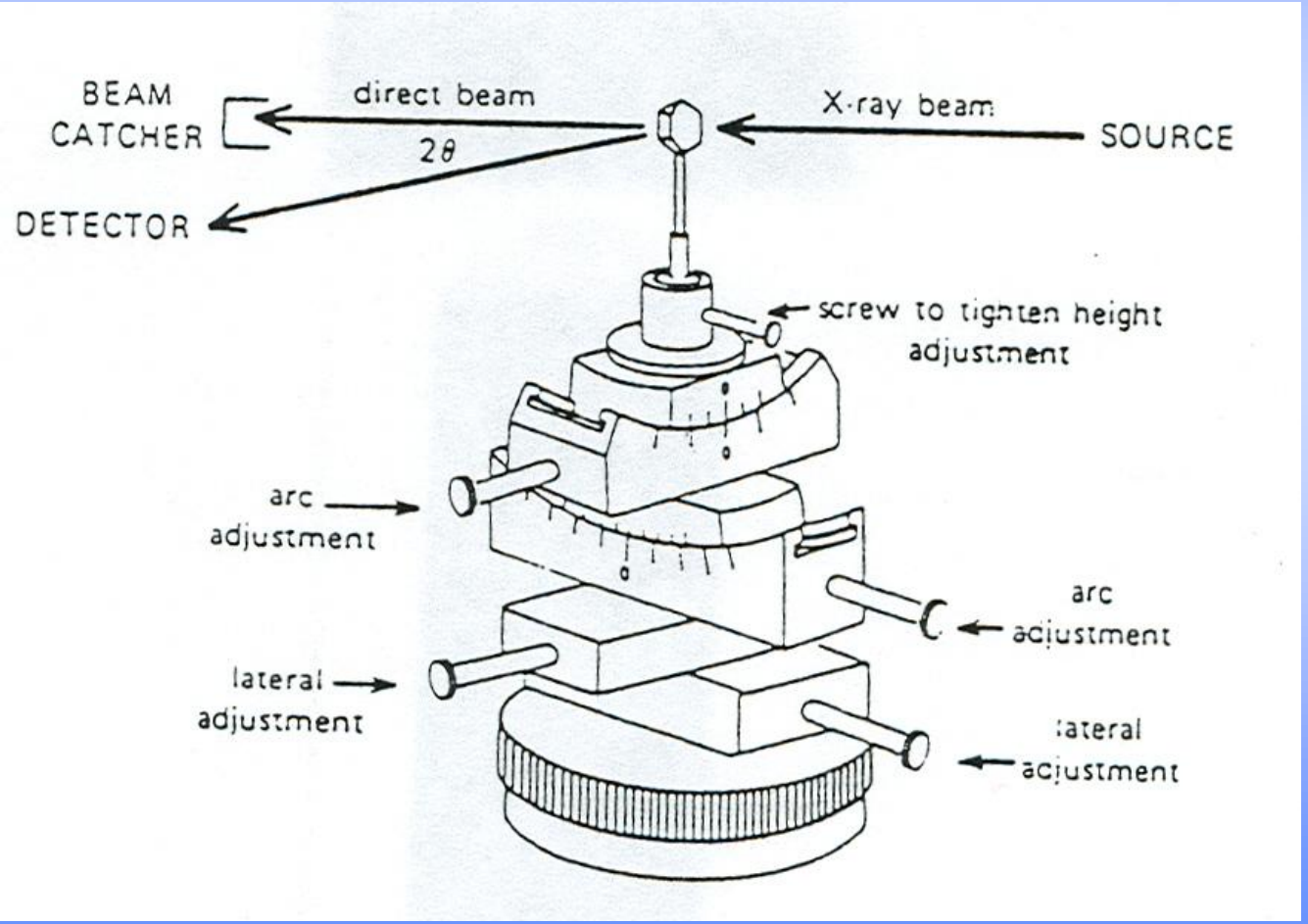
A) Assemble capillary, syringe and rubber tubing.

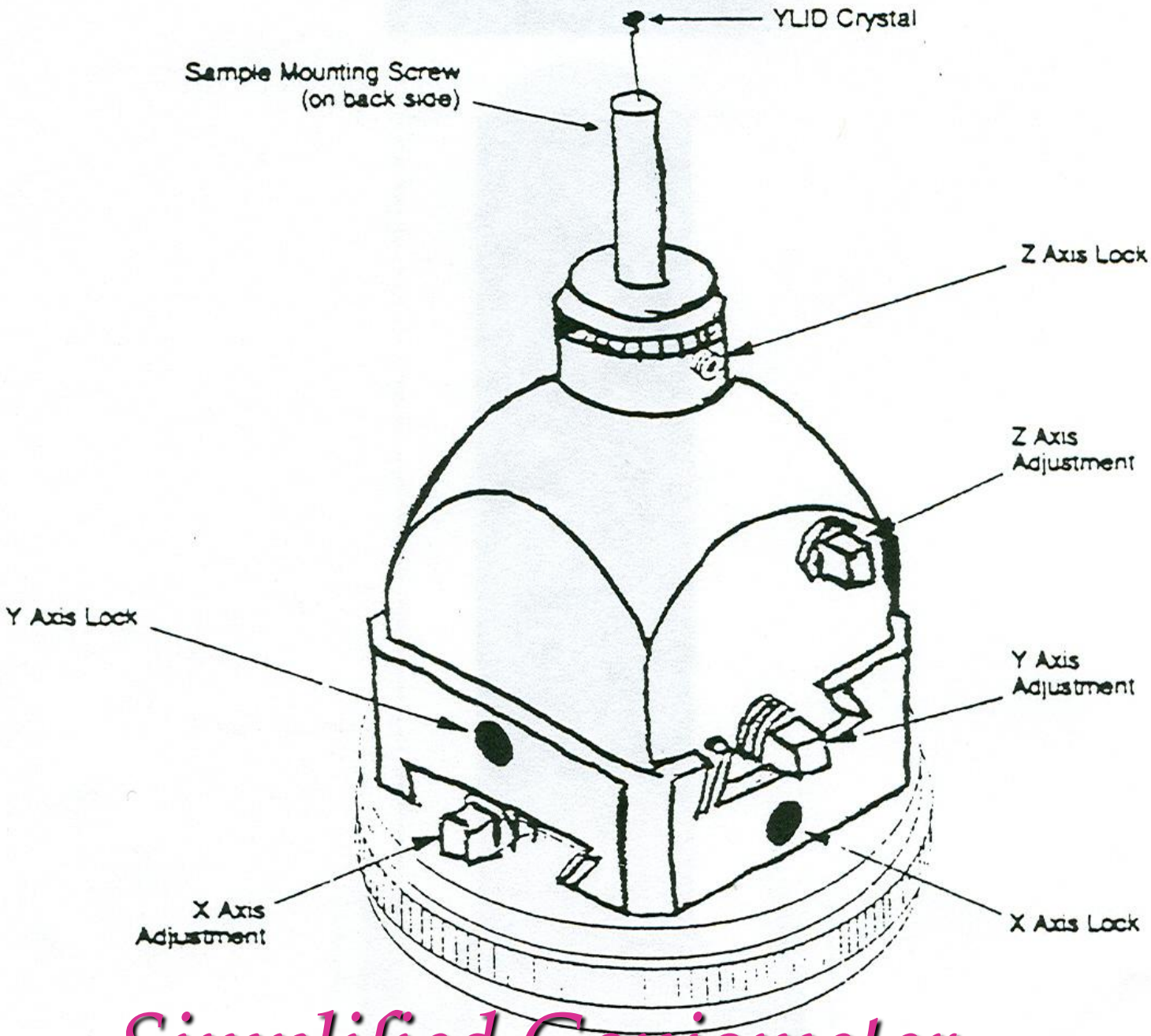
B) Under a low power-dissecting microscope, draw the crystal into the capillary.

C) Remove excess liquid with a paper wick.

D) Apply crystallization media to either side of the crystal (or both) in order to prevent desiccation of the sample. The capillary is broken at the location of the wax bead and the open ends are sealed with wax or vacuum grease.

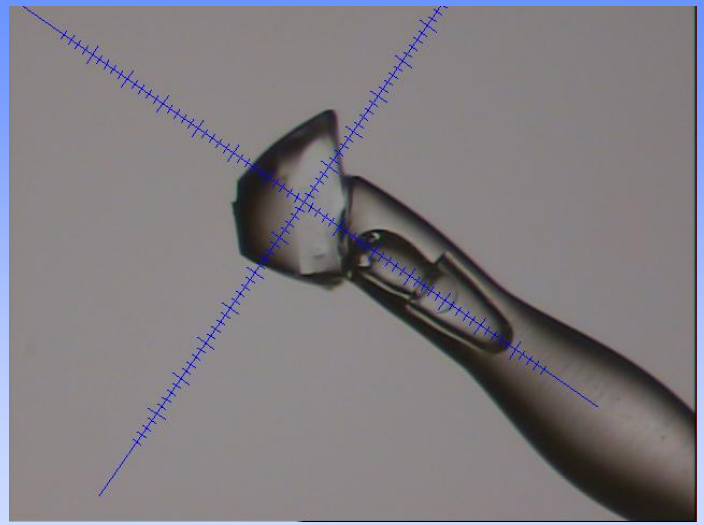
Crystal Needs to be in the center of the Beam

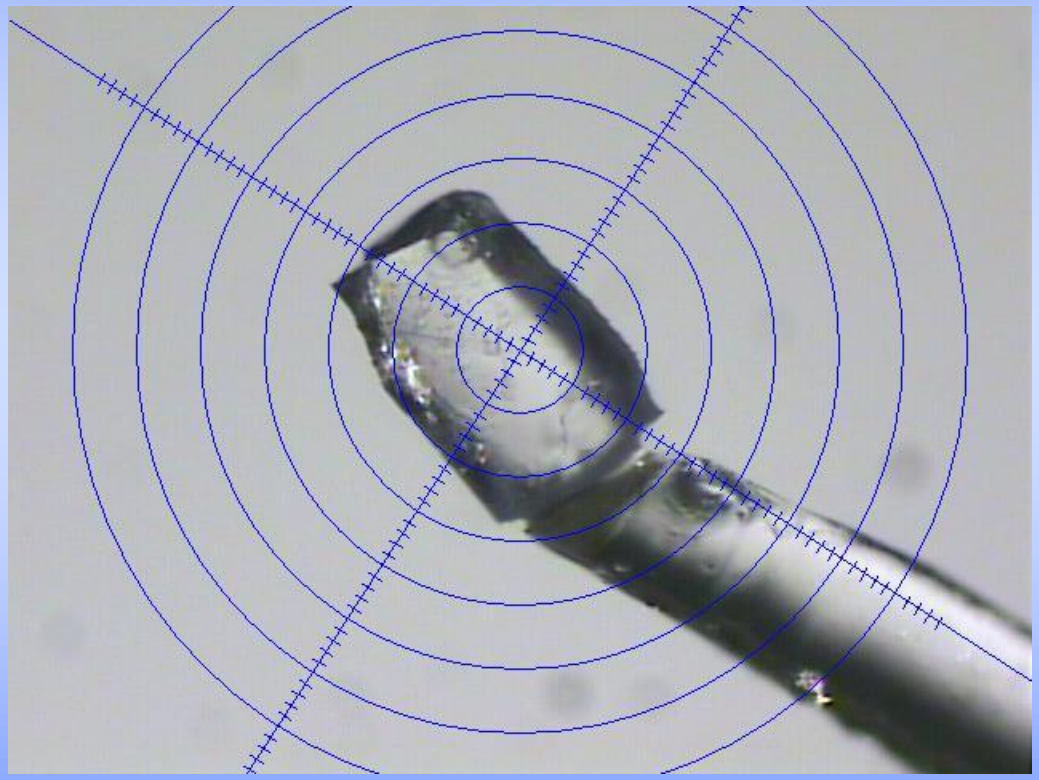


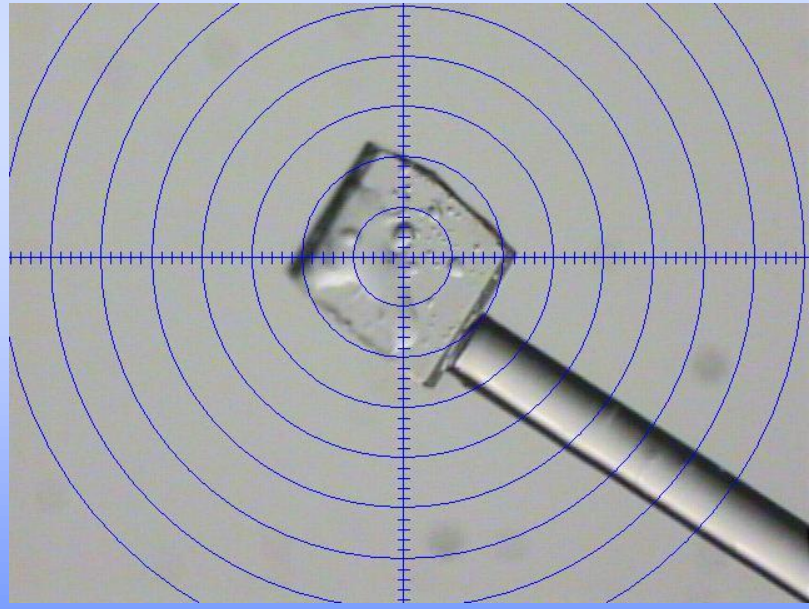


Simplified Goniometer

Centering



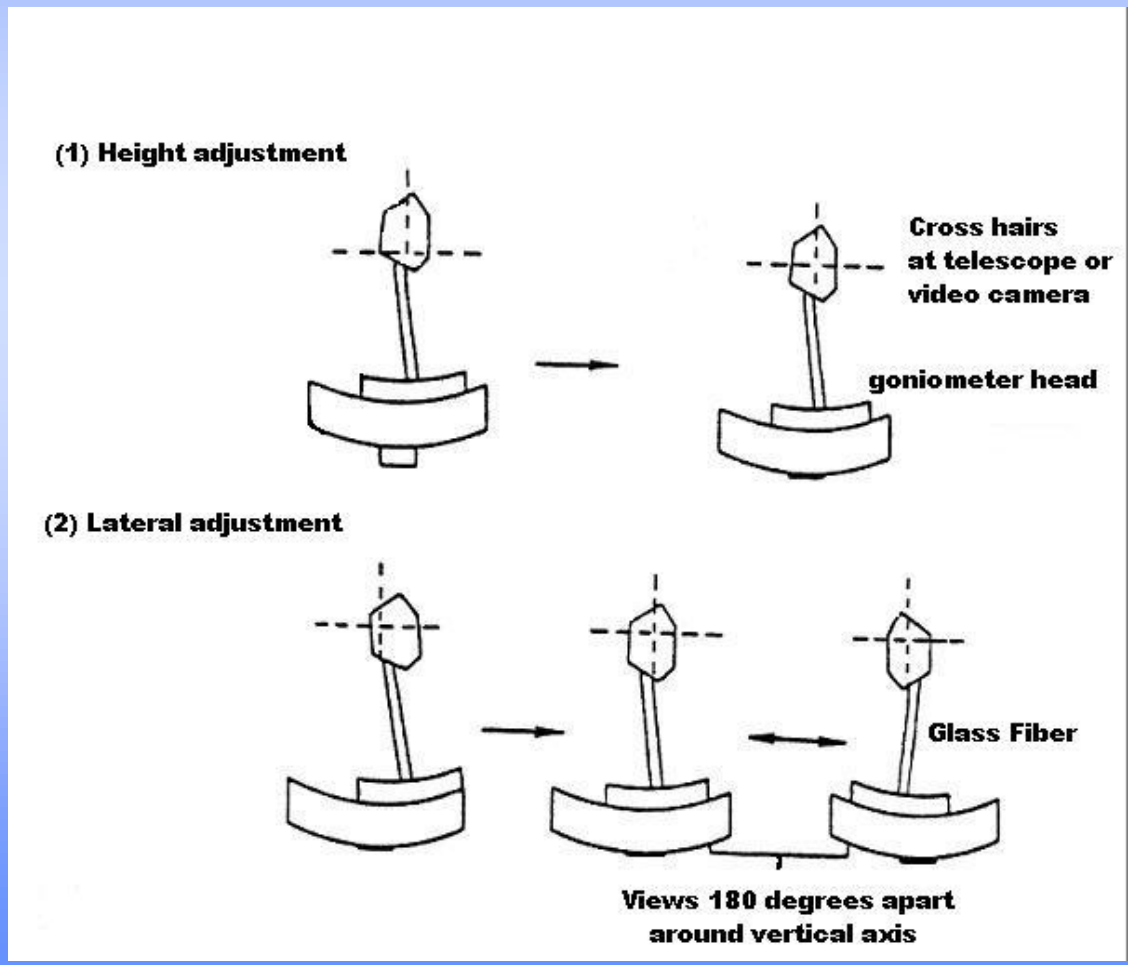




Sometimes the cross hairs do not represent the center of the x-ray beam and so rotation of the crystal by 180 degrees is done to facilitate alignment

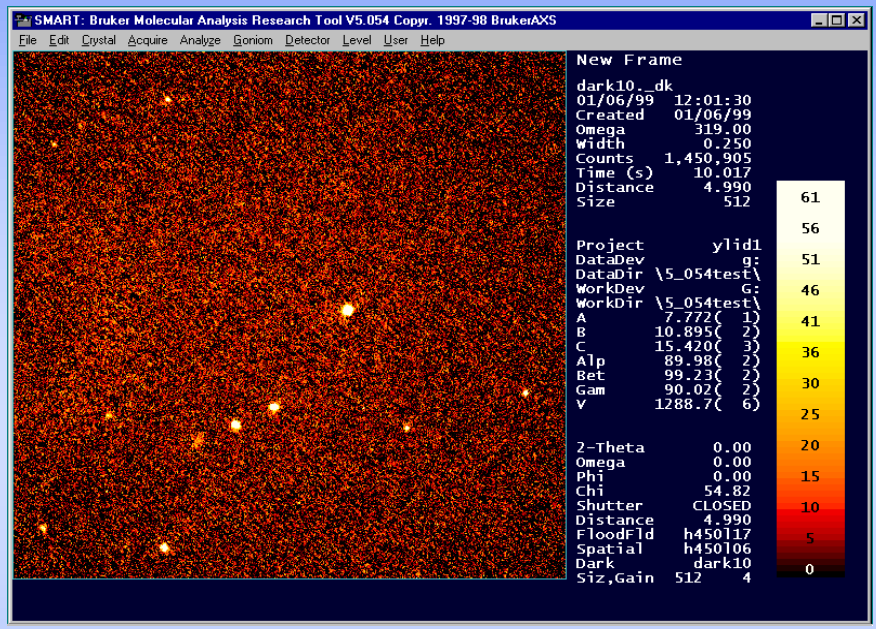
Rotation by 180 degrees

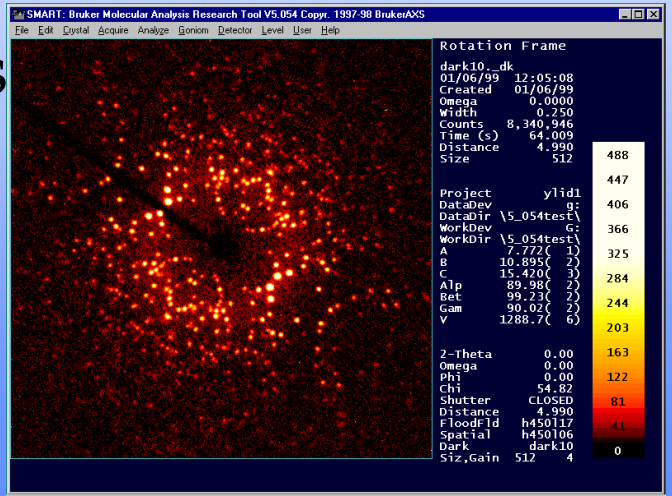
All modern instruments have a way that you rotate the sample with respect to the video camera or microscope, such that you can rotate a perpendicular view by 90 and 180 degrees. This allows you to center the crystal in the beam even if the center of the video camera or microscope is not correct.



The Key Question: Does it Diffract?

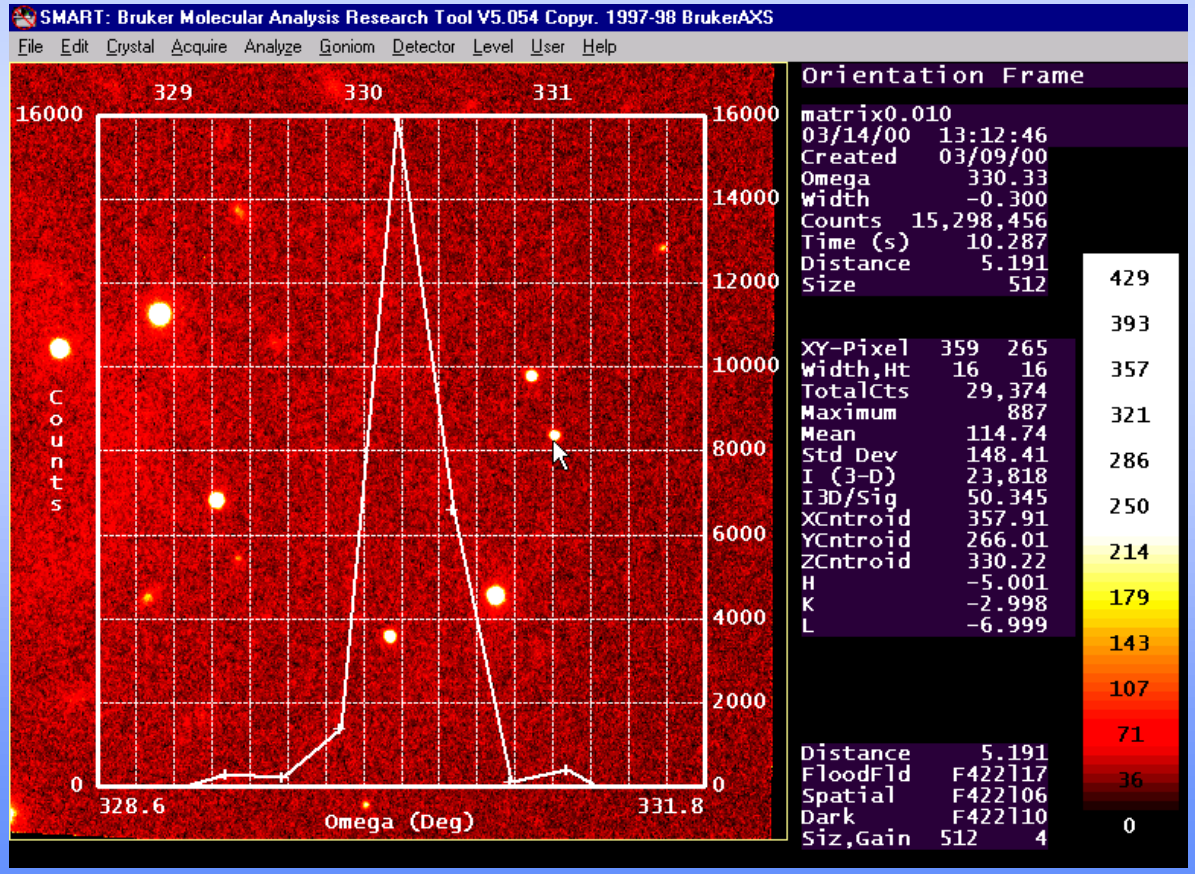
- Screening is very fast with CCD
- Run matrix at 10 second exposure and look for diffraction spots.
- Further out the better
- Spots should change positions over two or three frames
- Run Rotation Photo





Evaluate the Cell if given.

Look at Rocking Curve



Limitations to Crystallography.

- Requires single crystals (although PXRD using Reitveld method is possible)
- Crystal quality governs quality of results obtained.
- Only one crystal of the bulk material.