Growing X-ray Quality Crystals

Paul D. Boyle
Department of Chemistry
University of Western Ontario

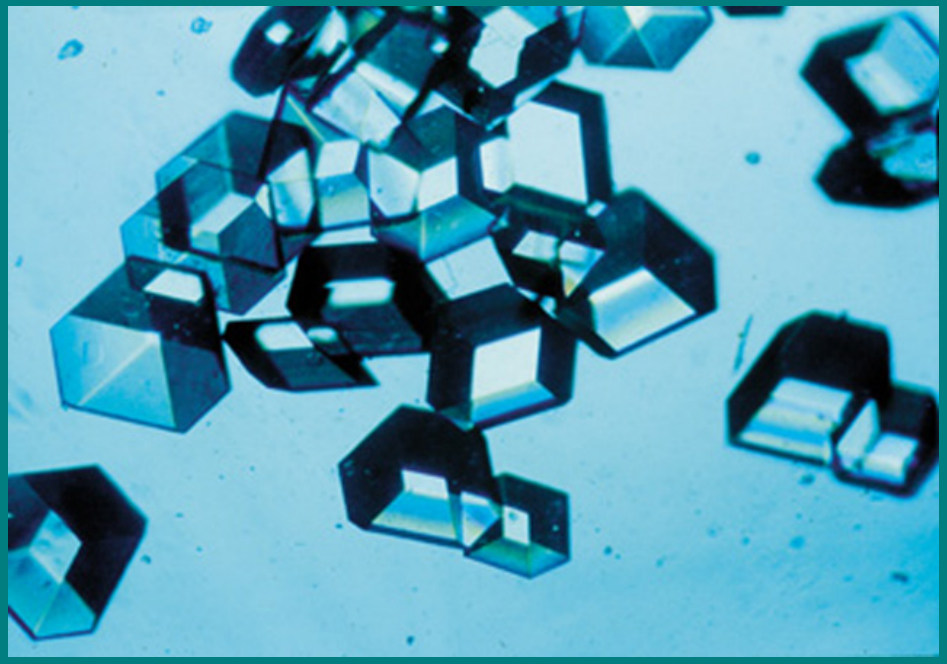
Important Caveats!

- X-ray Crystallography **does not** determine your compound's chemical composition
- It is a technique for determining atomic connectivity of a single crystal. Is your single crystal representative of the bulk composition?
- Samples submitted for X-ray crystallographic analysis need to have other characterization data (e.g. NMR, IR, MS, ESR, elemental analysis, etc.) to establish the chemical formula
- Chemical research should be bulletproof and the result of being thorough, paying attention to detail, and making sure every conclusion is backed with empirical evidence. Don't make assumptions or jumps in logic that your crystal structure is actually representative of your major reaction product.

Topics to be Covered

- The Objective: Getting “Good” Crystals
- Why do you need good crystals anyway?
- The Right Attitude toward crystal growing
- Factors affecting crystallization
- Crystal growing techniques
- Questions and Answers/Discussion

Objective: Getting Good Crystals

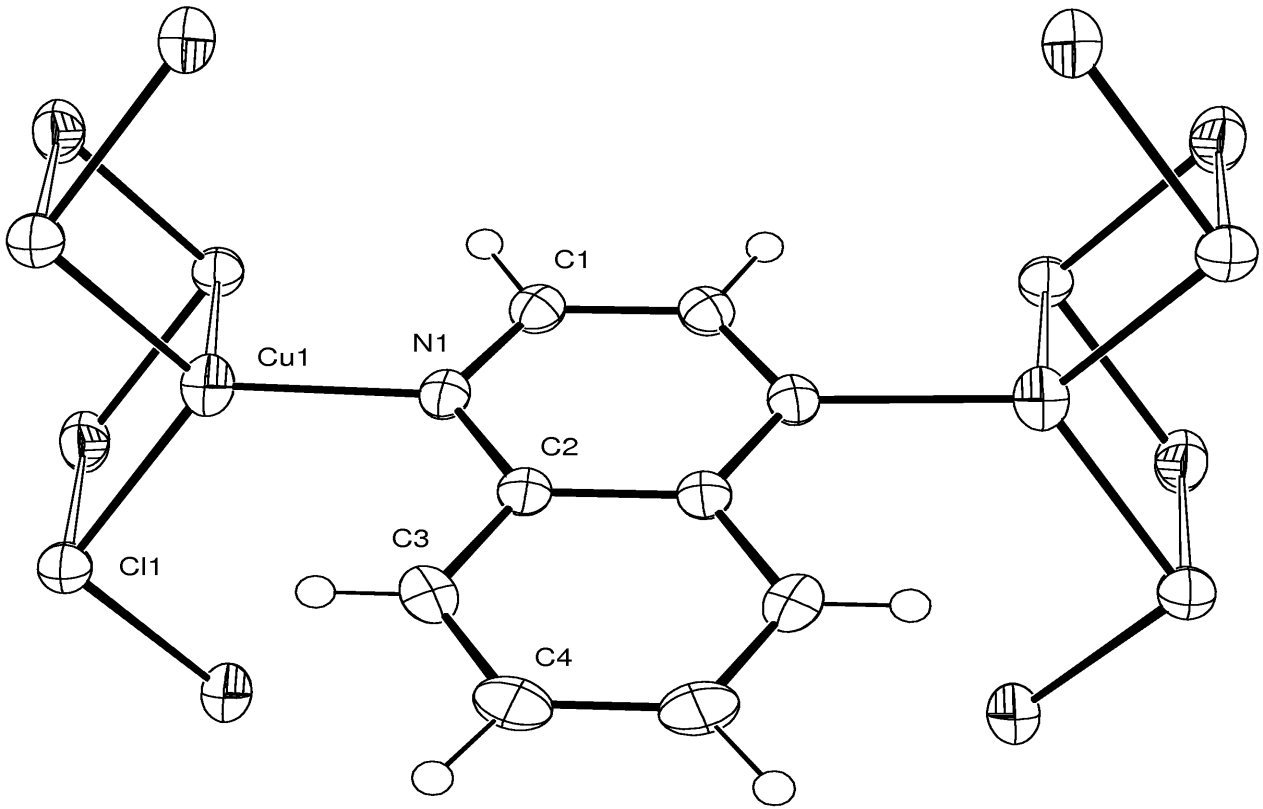


- A “good” crystal is:
- 0.1-0.3 mm in at least 2 of its dimensions
- exhibits a high degree of internal order as evidenced by the presence of an X-ray diffraction pattern
- Very often, but not always, shows regular faces and edges

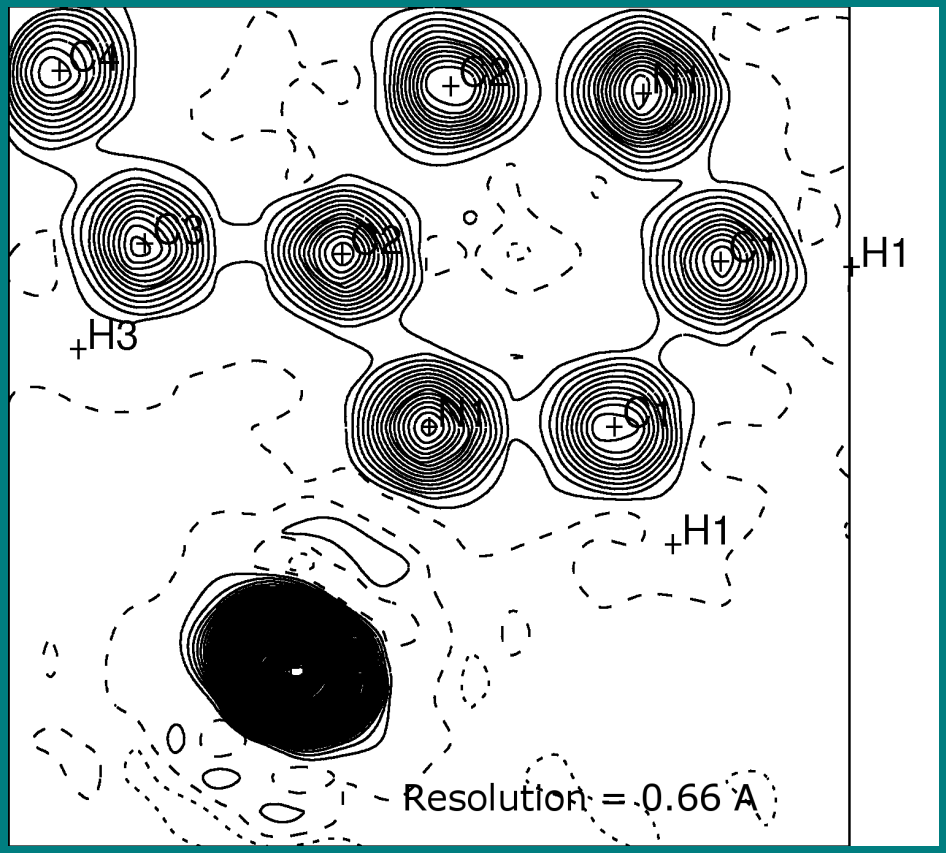
Why do you need good crystals anyway?

- Quality of sample is characterized by maximum diffraction angle (θ) -- also expressed in “resolution” (Å)
- The larger the max. diffraction angle, the higher the resolution and the greater number of data (which are necessary to adequately model the structure)
- Discerning individual atomic positions requires data resolution which is higher than chemically significant distances (e.g C=O ~= 1.2 Å)

The Effect of Limiting the Resolution of the Data

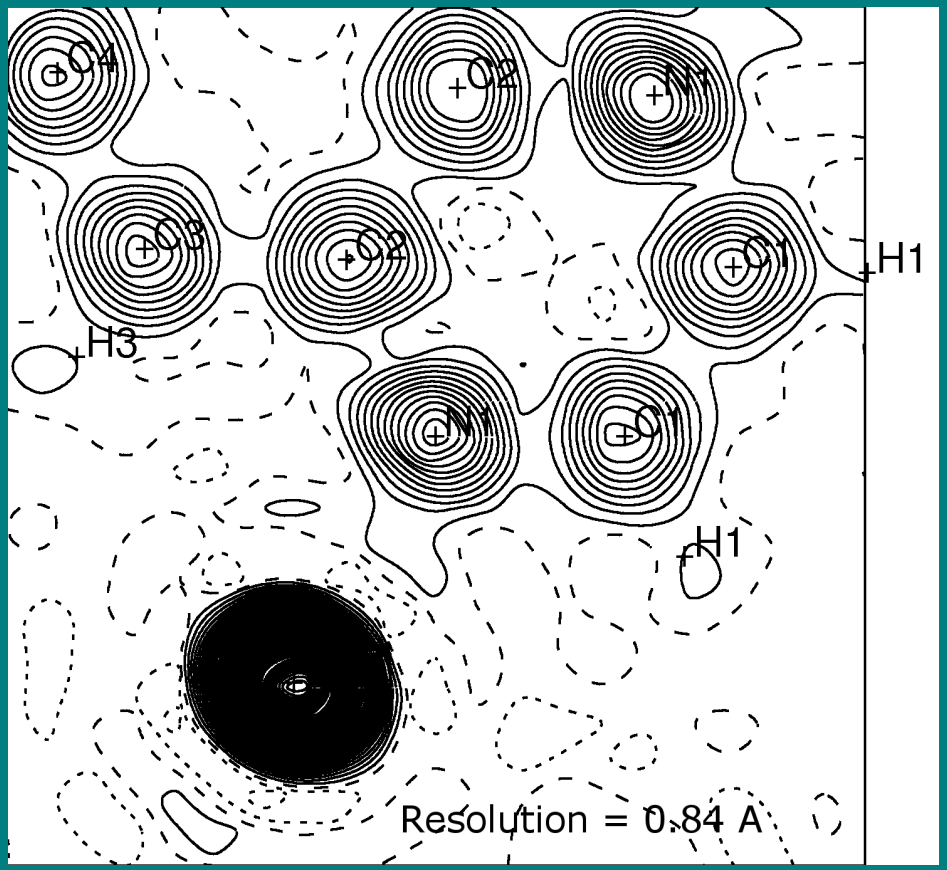


Effect of Limiting Resolution of the Data



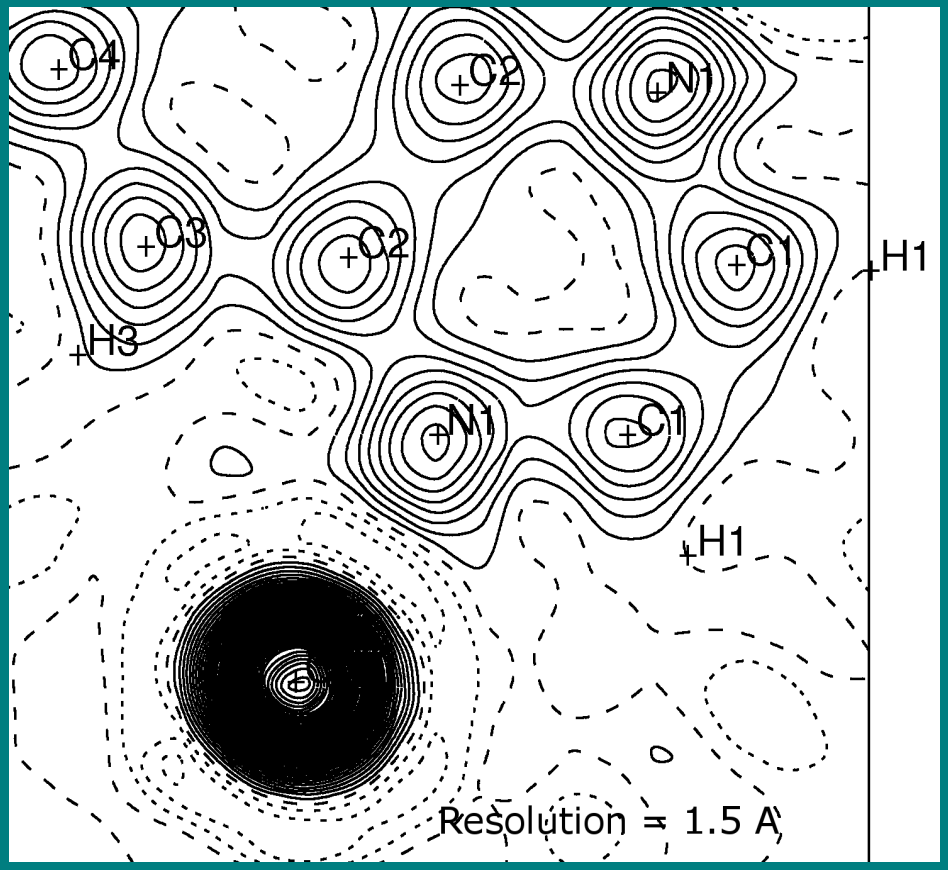
- Electron Density Map using all available data (θ_{max} = 32.35°)
- Resolution = 0.66 Å
- All atomic positions are easily resolved

Limiting the Resolution of the Data



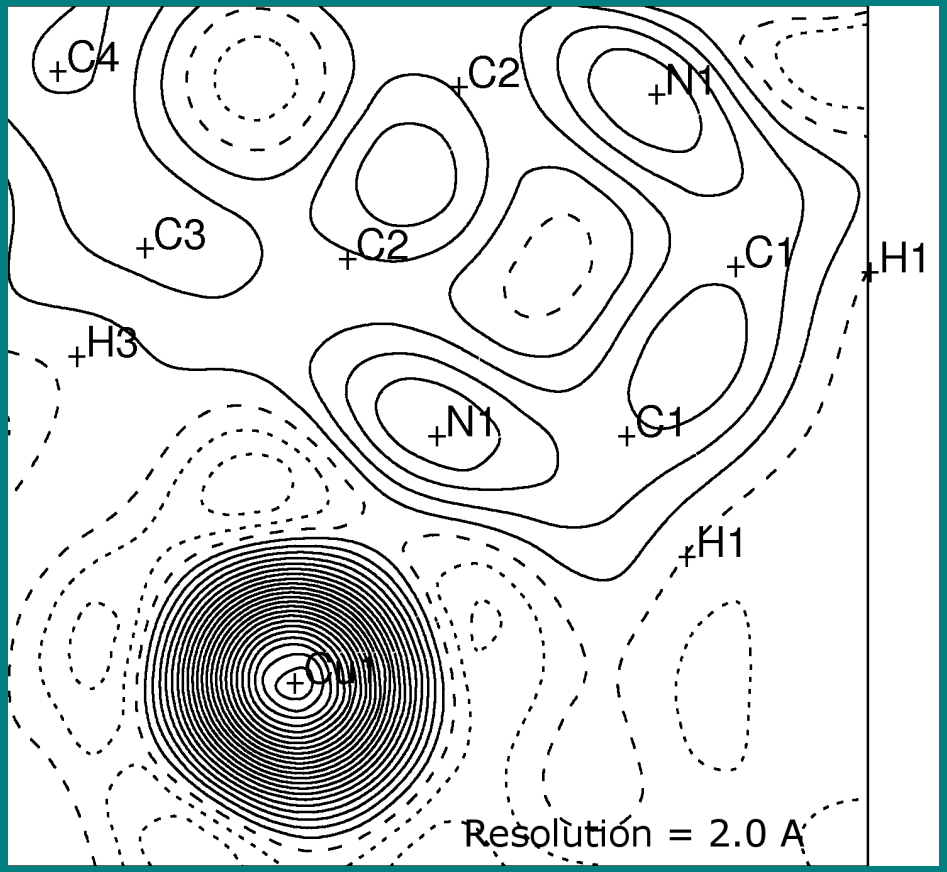
- Limited θ_{max} = 25.0°
- Resolution = 0.84 Å
- Peaks are beginning to flatten out
- Atomic positions are still easily resolvable
- IUCr recommended <u>minimum</u> resolution

Limiting the Resolution of the Data



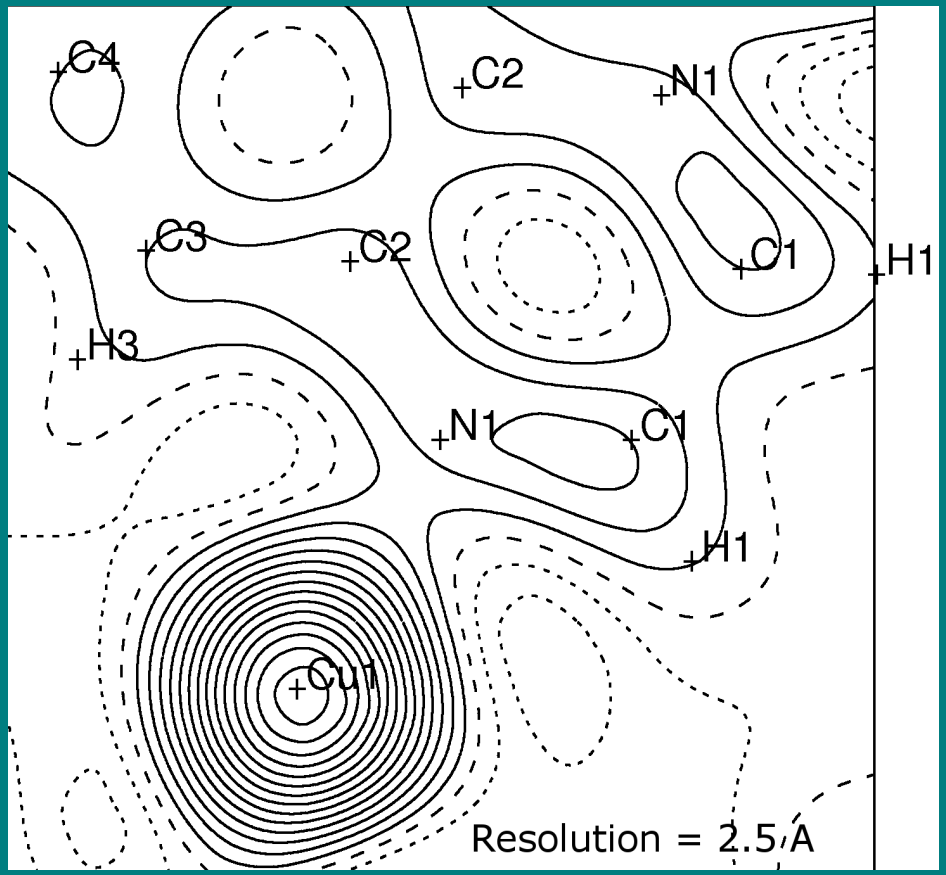
- Limited θ_{max} = 19.47°
- Resolution = 1.5 Å
- Peaks start to “melt” into each other
- Individual atomic positions are still resolvable

Limiting the Resolution of the Data



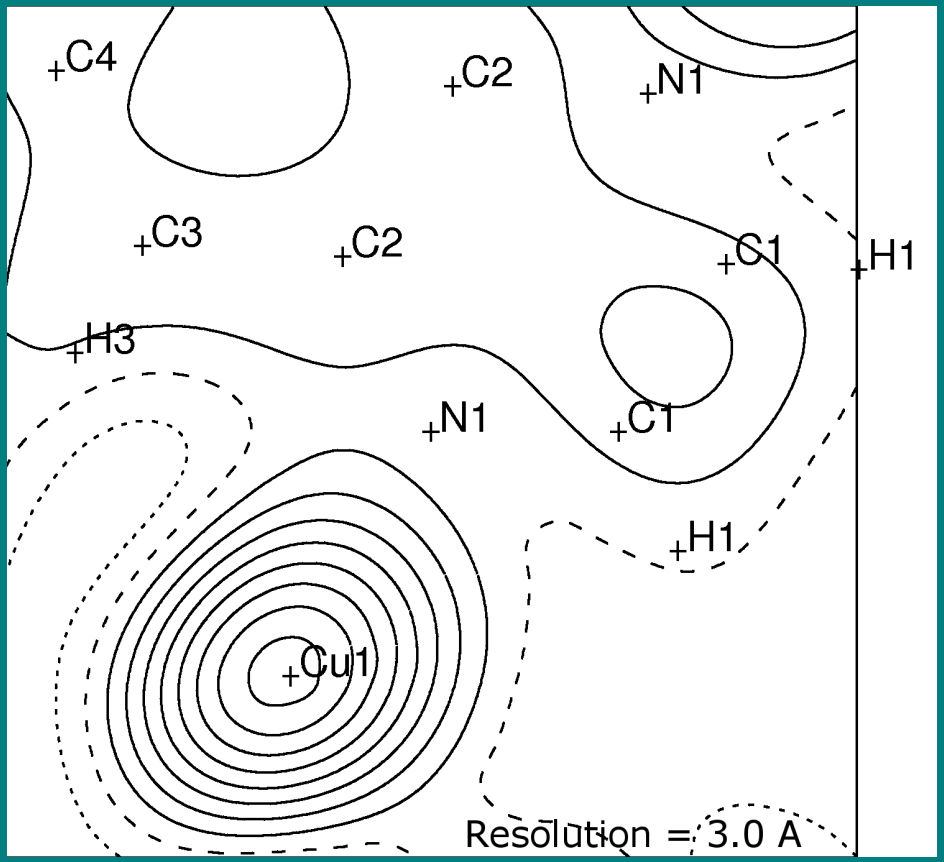
- Limited θ_{max} = 14.48°
- Resolution = 2.0 Å
- Metal position resolvable
- Only gross shape of organic ligand evident
- Peak positions for ligand have shifted away from true atomic positions

Limiting the Resolution of the Data



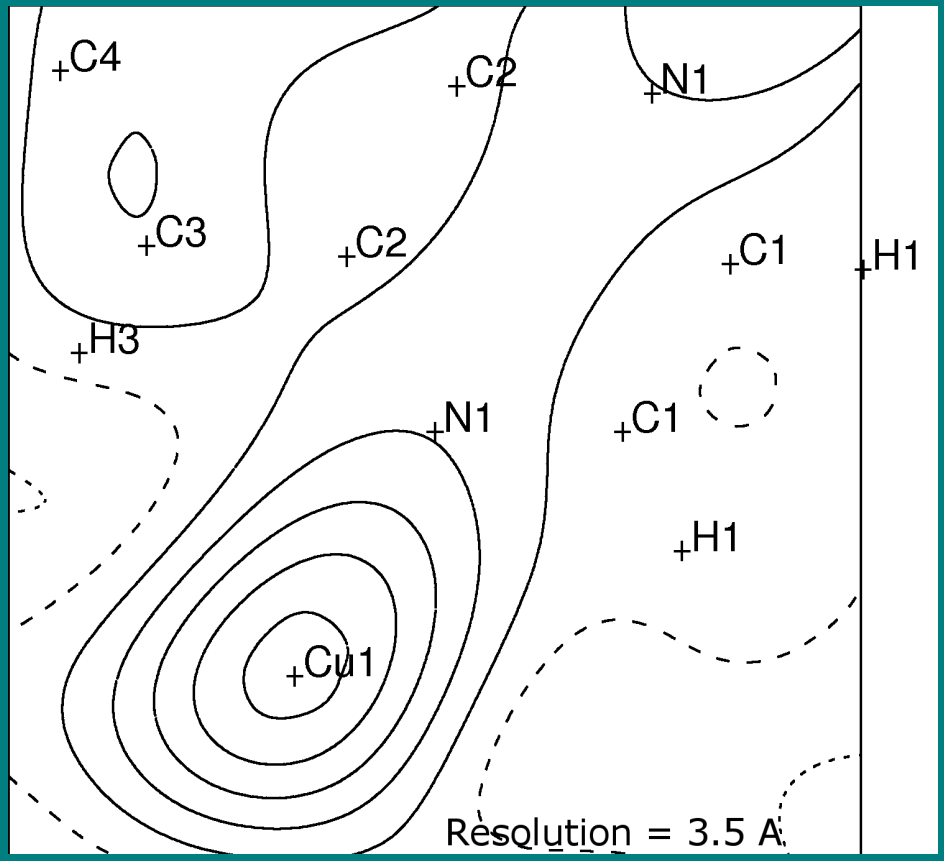
- Limited θ_{max} = 11.54°
- Resolution = 2.5 Å
- Metal position still discernible
- Individual atomic positions for ligand have been lost
- Lost of chemically useful information

Limiting the Resolution of the Data



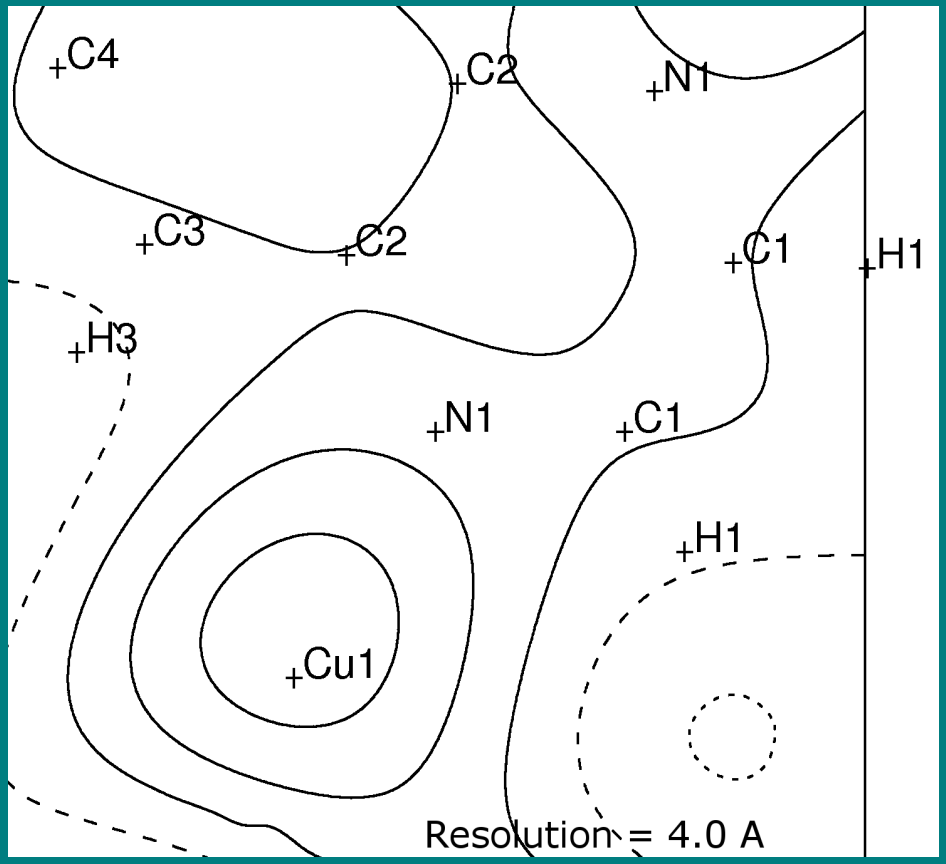
- Limited θ_{max} = 9.59°
- Resolution = 3.0 Å
- Only metal position is discernible
- Ligand is completely “washed out”

Limiting the Resolution of the Data



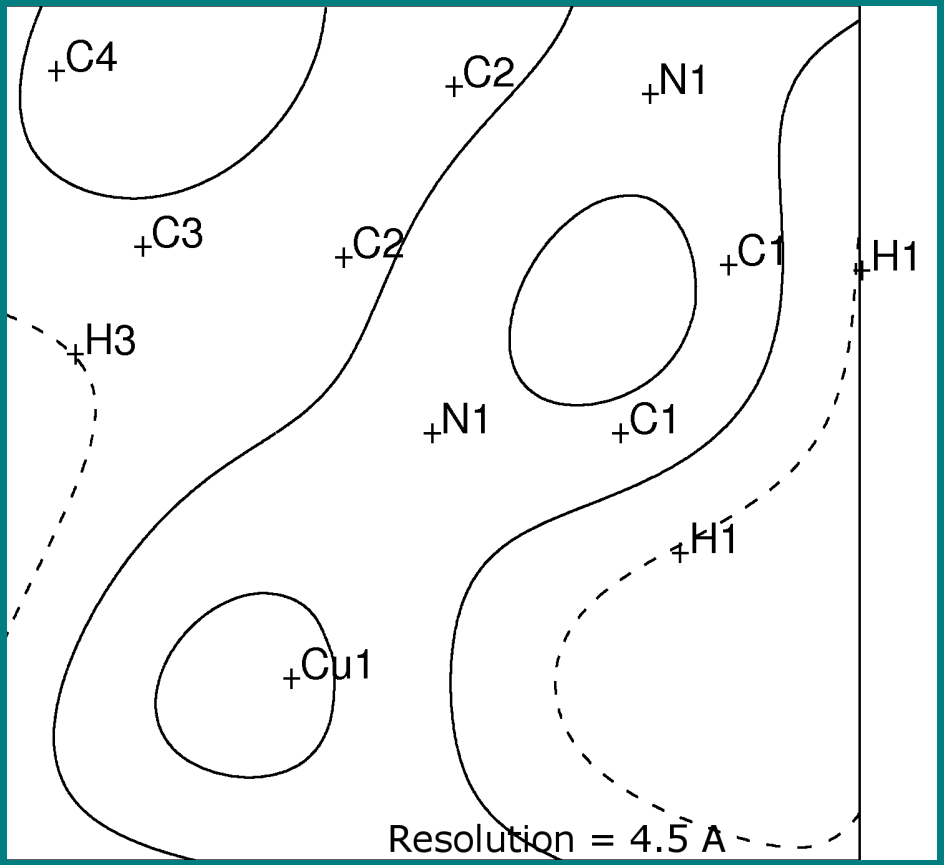
- Limited θ_{max} = 8.21°
- Resolution = 3.5 Å
- Metal electron density is “bleeding” into the traces of the ligand's electron density

Limiting the Resolution of the Data



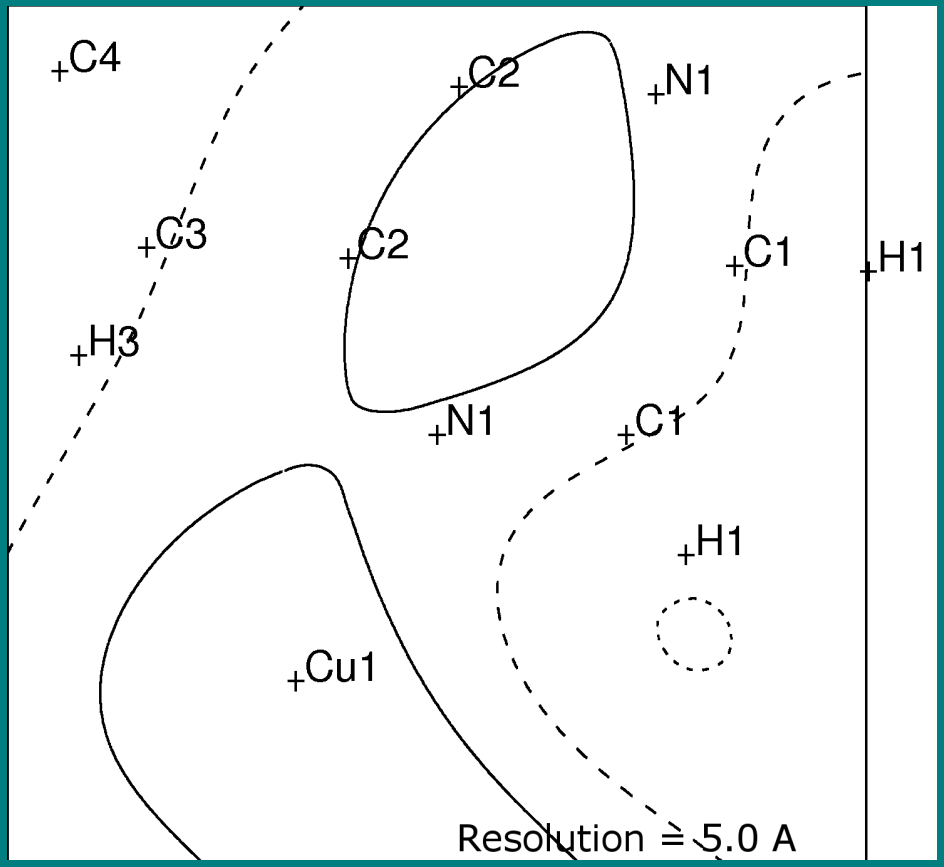
- Limited θ_{max} = 7.18°
- Resolution = 4.0 Å
- Metal position is only barely above background
- No trace of ligand

Limiting the Resolution of the Data



- Limited θ_{max} = 6.37°
- Resolution = 4.5 Å
- Metal position cannot be differentiated from noise

Limiting the Resolution of the Data



- Limited θ_{max} = 5.74°
- Resolution = 5.0 Å
- Noise

The Right Attitude toward Crystal Growing for X-ray Analysis

- Growing X-ray quality crystals requires care and attention to detail
- Don't treat crystal growing in an offhand or casual way (forget what you learned in undergraduate organic chemistry labs)
- Treat it like its own miniature research project
- If you've spent weeks or months doing your synthesis, why assume finding the correct crystallization conditions will just take a few hours?
- Don't try to skimp on the amount of material when growing crystals

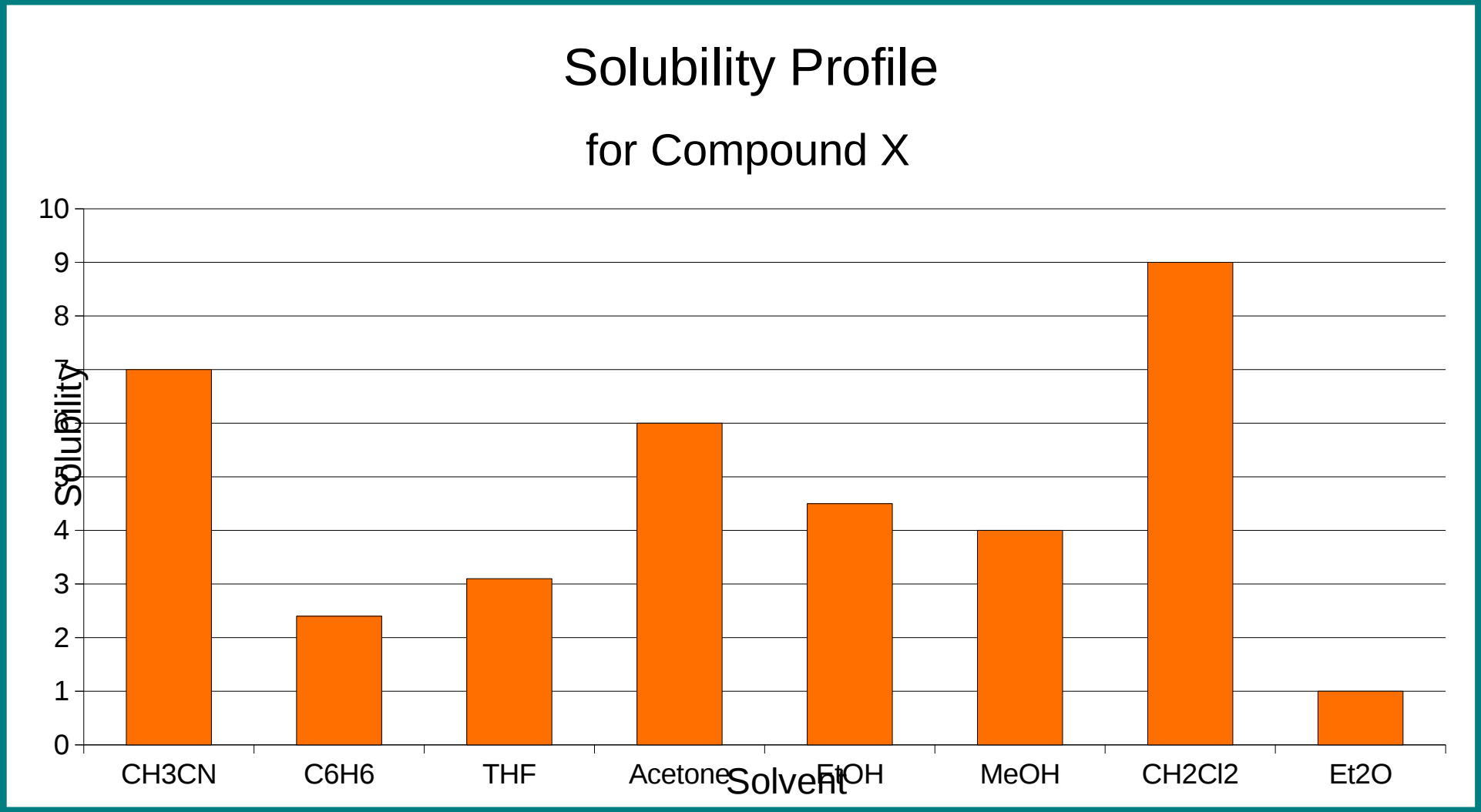
General Approach to Growing X-ray Quality Crystals

- Purify your compound (using conventional crystallization and/or other purification steps)
- Consider the empirically established physical properties of your compound – sensitivities, thermal stability, etc.
- Develop a **solubility profile** of your compound
- Use **CLEAN** glassware as crystal growing vessels
- Set up crystal growing attempts in **parallel** utilizing different conditions

Purify Your Compound

- Impure samples do not recrystallize as well as pure samples
- Recrystallization minimizes the presence of foreign insoluble material which increases the number of nucleating sites
- Successive crystallizations purify the compound
- Always use recrystallized material when setting up a crystal growing attempt
- Use NMR to establish that your compound hasn't decomposed during crystallization

Solubility Profile



Use **CLEAN** Glassware

- Clean glassware should sheet water uniformly
- Use KOH/EtOH bath or *aqua regia* to initially clean glassware and rinse
- Follow by soap & water washing
- Follow by acetone or MeOH rinse
- Oven drying

Another thing about Glassware

- Before setting up a crystal growing attempt think about how the crystals will be handled
- Crystals will need to extracted from the vessel without damage
- Therefore, pick a suitable crystal growing vessel
  - **BAD:** Round bottom flasks of any size
  - **BAD:** Small aperture vials (too small for spatula)
  - **BAD:** Screw top vials (shoulder causes difficulty)
  - **GOOD:** NMR tubes
  - **GOOD:** Small test tubes
  - **GOOD:** Vials without shoulders

Parallel Crystal Growing Attempts

- Combine knowledge of solubility profile with crystal growing techniques
- Set-up simultaneous crystal growing experiments

Factors Affecting Crystallization

- **Solvent** – moderate solubility is best. Supersaturation leads to sudden precipitation and smaller crystal size
- **Nucleation** – fewer nucleation sites are better. Too many nucleation sites (i.e. dust, hairs, etc.) lower the average crystal size
- **Mechanics** – mechanical disturbances are **bad**.
- **Time** – faster crystallization is not as good as slow crystallization. Faster crystallization higher chance of lower quality crystals

Solvent Considerations

- Moderate solubility is best (avoid supersaturation)
- Like dissolves like
- Hydrogen bonding can help or hinder crystallization. Experiment!
- Presence of benzene can help crystal growth
- Avoid highly volatile solvents
- Avoid long chain alkyl solvents, they can be significantly disordered in crystals. Choose solvents with “rigid geometries” (e.g. toluene)

Nucleation & Growth

- Crystals initially form via “nucleating events”
- After a crystallite has nucleated it must grow
- Nucleation sites are necessary, but ...
- Excess nucleation sites cause smaller average crystal size
- Ambient dust, filter paper fibers, hair, broken off pipette tips all provide opportunities for nucleation – take steps to remove them.

Mechanics (Crystal Growth)

- Crystals grow by the ordered deposition of the solute molecules onto the surface of a pre-existing crystal
- Crystal growth is facilitated by the environment changing slowly over time.
- Keep crystal growth vessels away from sources of mechanical agitation (e.g. vibrations)
- Set-up away from vacuum pumps, rotovaps, hoods, doors, drawers, and so on
- Leave samples alone for 1 week, don't “check in” with it. Your crystals are **not** lonely.

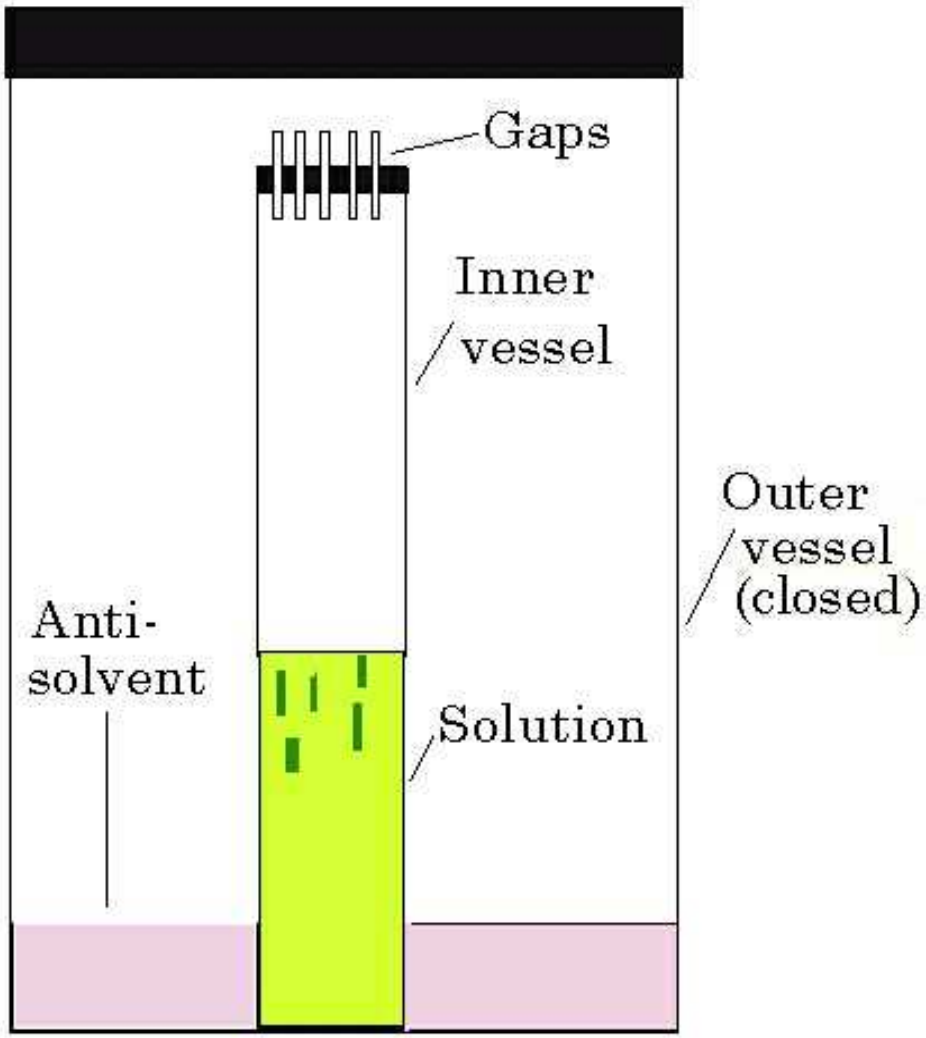
Time

- Quality crystals grow best over time in near equilibrium conditions
- The longer the time, the better the crystals
- Larger crystals tend to grow at the expense of smaller crystals
- Patience, patience, patience

Crystal Growing Techniques

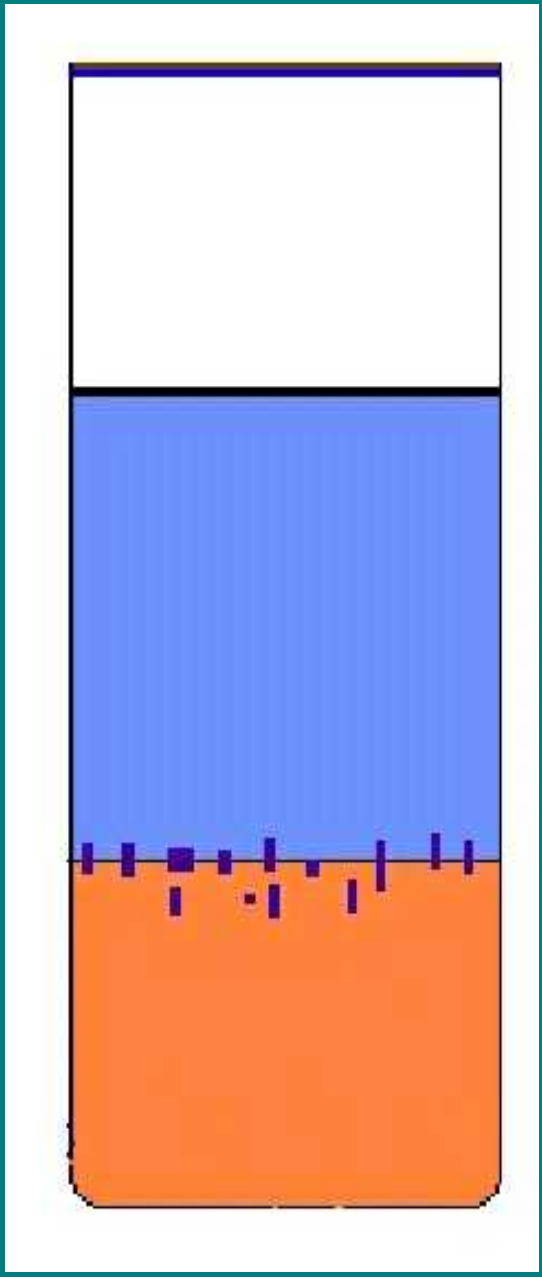
- **Slow Evaporation:** simplest to set up. Has drawbacks: solute can “oil out”, crystals stick to sides of vessel making them difficult to extract from vessel without breaking them..
- **Slow Cooling:** Soluble when hot, insoluble when cool. Use Dewar to slow the cooling process.
- **Variations:** use binary or tertiary solvent mixtures. Use solvent with similar b.p's and other properties. Document the percentages of each solvent component!

Vapour Diffusion



- dissolve solute in solvent (green)
- precipitating “anti-solvent” (pink)
- pink should be more volatile than green
- Don't let sides of small vessel touch vertical surface of outer vessel (prevent capillary action)

Solvent Diffusion

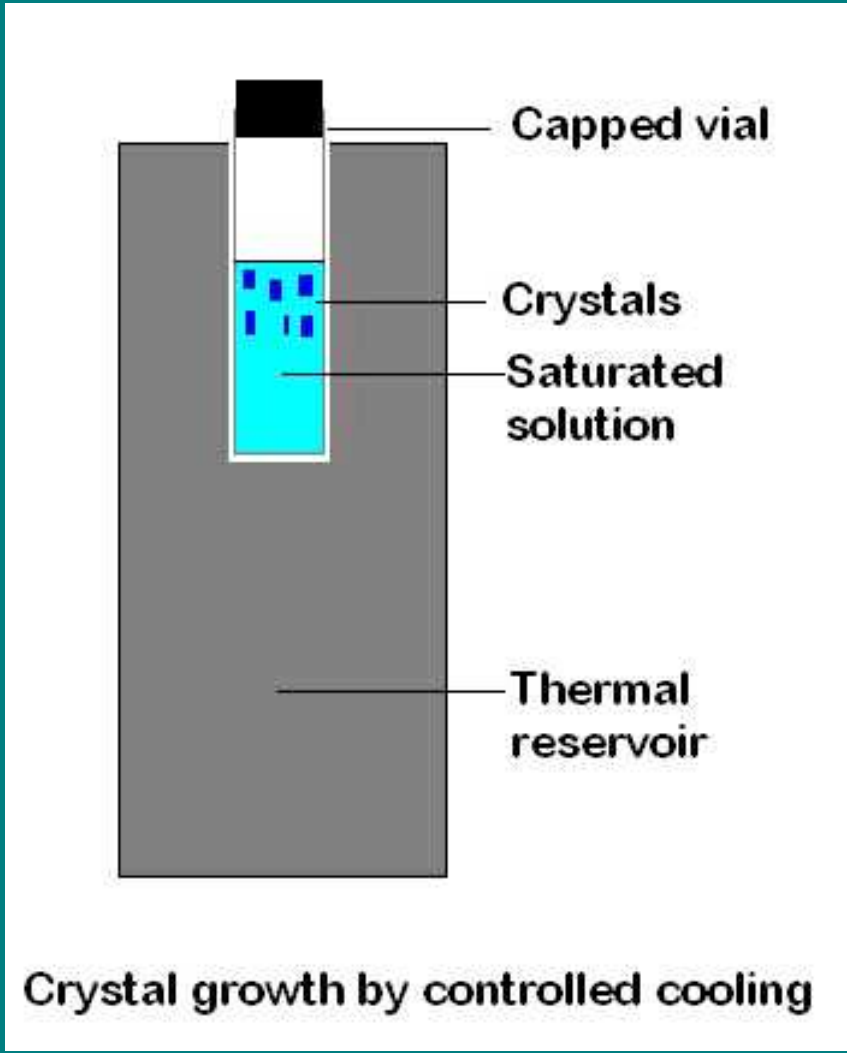


- Good for milligram amounts
- Use NMR tube for best results
- Fill soluble more dense solvent on bottom with your solute.
- Fill the rest of tube with less dense precipitant solvent
- E.g. CH_2Cl_2/Et_2O

Reactant Diffusion

- Set-up similar to solvent diffusion except that reactants are in different layers
- Good for milligram amounts
- Good for completely insoluble products which never go back into solution after being formed
- Consider using a 3rd “middle layer” solvent to mediate the reactant concentrations

Slow Cooling



Crystal growth by controlled cooling

- Heat water to just below the boiling point of the solvent you plan to use
- Create a saturated solution of your compound in boiling solvent
- Transfer to solution to a test tube and cap
- Fill Dewar with hot water
- Place test tube into the Dewar and cap Dewar
- Let stand several days

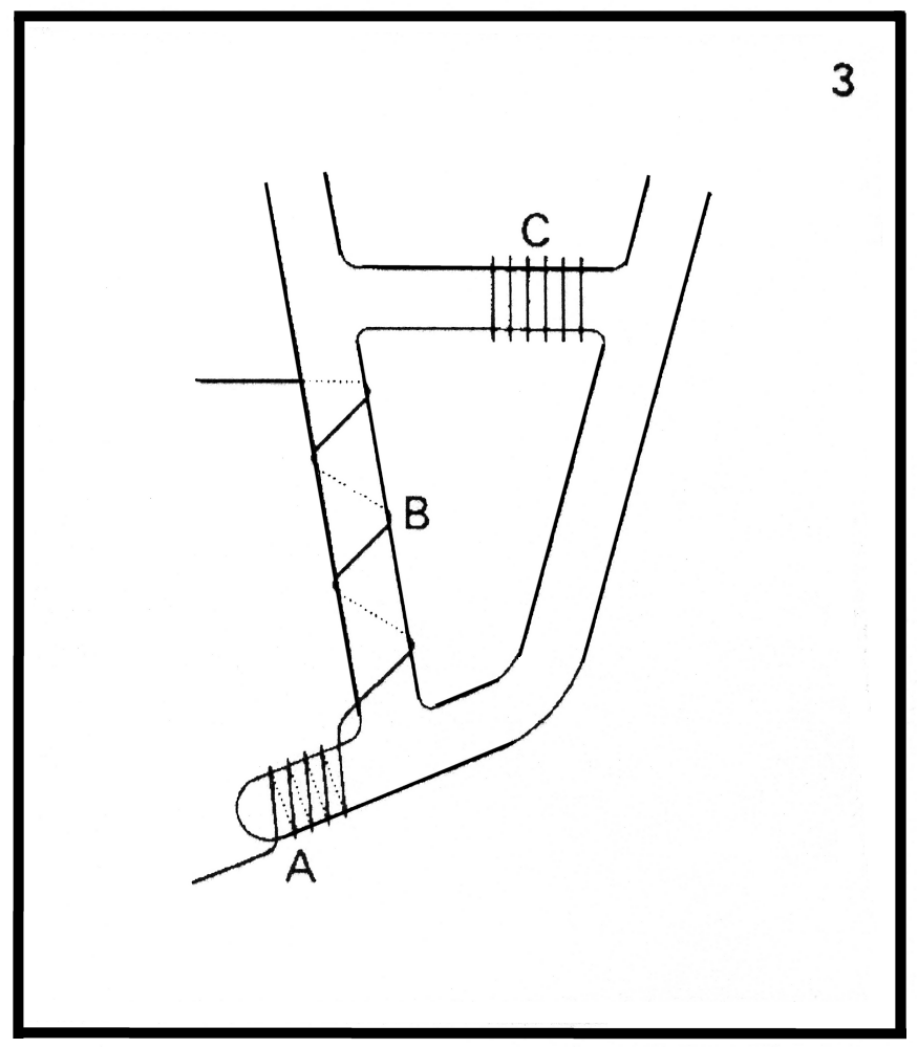
Convection (Principles)

- Good for insoluble or sparingly soluble compounds
- Create a thermal gradient in the crystal growing vessel
- Solvent becomes saturated in “warm” region and deposits material in “cool” region where nucleation and crystal growth can occur. Cyclic current allows continual replenishing of the solute
- Velocity of convection current is proportional to the magnitude of thermal gradient.
- Take care that the gradient isn't too large – too high velocity inhibits crystal growth

Convection (Easy Way)

- Local cooling – simple to set up
- Take flat bottomed crystal growing dish and set up like slow evaporation
- Place vessel so that one **side** is against a heat sink, e.g. an outside window (in Winter at least)
- Placing crystal growing dish on a cool surface will **not** cause convection.

H. Hope's Convection Device

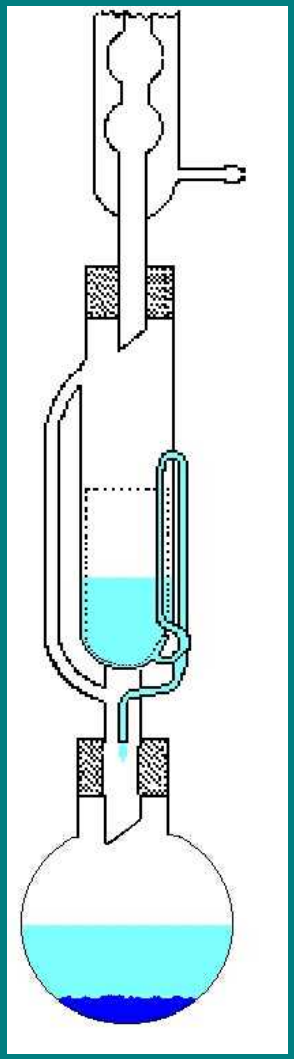


- Specialty glassware can be fabricated
- This device designed by Håkon Hope (*J. Appl. Cryst*. **1971**, *4*, 333)
- Nichrome wire is used for the main [A] and subsidiary [B] heating elements.
- The solvent is cooled at point [C] using Cu wire or thin plastic tubing as a heat sink.

Thiele Tube Convection Device

- If you don't want to fabricate a special piece of glassware – improvise!
- Fill Thiele tube with solvent.
- Wrap nichrome wire around the bottom side arm and attach to Variac
- Place solute is small container just below top side entrance
- Apply heat

Soxhlet Extraction for Low Solubility Compounds

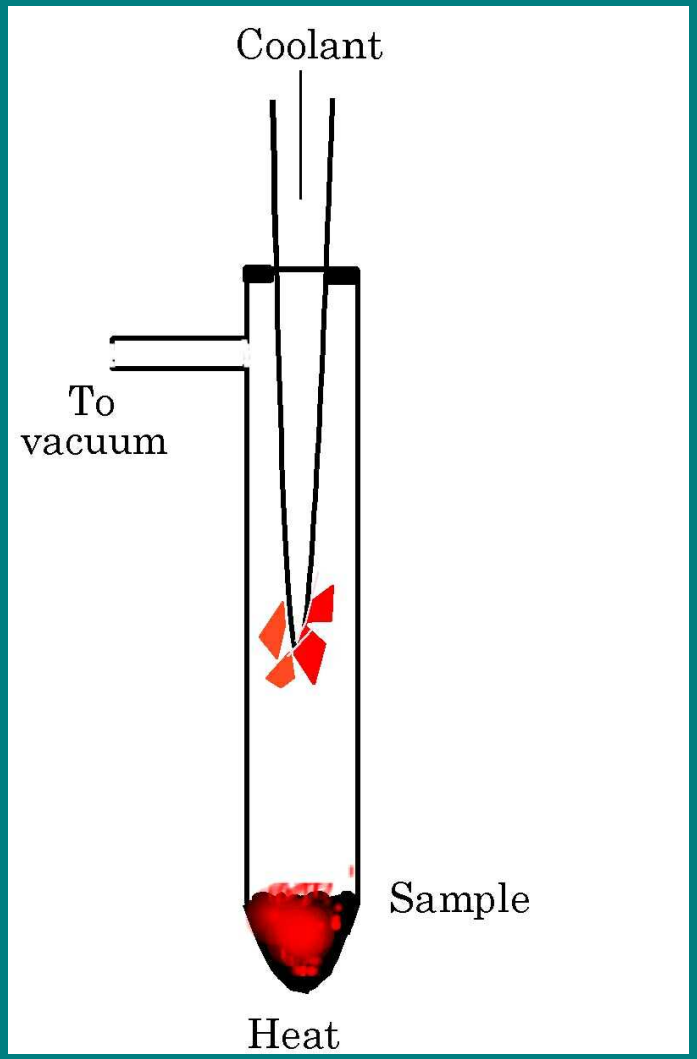


- Soxhlet extraction is normally used for separations. However, it can be used for crystal growing of thermally stable, sparingly soluble materials
- Place your solute in the sample thimble
- Start the solvent refluxing
- Crystals grow in the solvent reservoir

Sublimation (1)

- Gas to solid phase crystal growth
- Compound needs to be thermally stable
- Can be easy to set up – vacuum sealed tube of material placed in oven for several days/weeks
- Or more complicated – material packed in tube followed by glass wool. Place under active or static vacuum and set-up thermal gradient by heating the loaded end of the tube. Place Cu pipe around tube to create thermal gradient.

Sublimation (2)



- Good for volatile air-sensitive materials
- Specialty glassware is available
- Use minimal heat to sublime slowly
- Use small amounts of material

Co-Crystallants

- Sometimes two (or more) different compounds “co-crystallize”. Most commonly, this is a solvent molecule
- Triphenylphosphine oxide has been used as a co-crystallant for both inorganic and organic compounds

Chemical Modification

- For ionic compounds, change the counterion to change the solubility and other characteristics of your compound
- Ions of similar sizes tend to pack together better
- Use counterions with rigid geometries e.g. triflate, BPh_4^-, Me_4N^+, $(Ph_3P)_2N^+$
- Tend to disorder: Et_4N^+, Bu_4N^+, BF_4^-, PF_6^-
- Make sure counterion **does not react** with your compound!

Chemical Modification (Ionization of Neutral Compounds)

- If your compound is neutral and has proton acceptor or donor groups, consider ionizing the compound
- The ionic form may take advantage of hydrogen bonding to give better crystals
- Counterions can be changed to optimize crystal growth
- This will change your compound, but if you are only interested in confirming a structure, and not in detailed electronic properties, this shouldn't be a problem.

Online Resouces

- Use google (around 111,000 hits for 'X-ray “crystal growing”'), the knowledge is out there!
- http://xray.chem.uwo.ca/Guides.html
- http://www.nottingham.ac.uk/~pczajb2/growcrys.htm
- http://www.cryst.chem.uu.nl/lutz/growing/growing.html
- http://xrayweb.chem.ou.edu/notes/xtalgrow.html

Conclusion

- The quality and meaningfulness of your X-ray results is directly dependent on the quality of your sample crystal
- You can get information from a bad crystal structure, but it will be difficult to publish and makes for a weaker manuscript
- Take crystal growing as a serious part of your research project – spend the time and effort to be successful.
- There are many solvents and crystal growing techniques available – use them.

Acknowledgments

- Sandy Blake and Doug Powell for allowing me to use images from their crystal growing guides
- Clarence Pfluger, Tony Linden, Sandy Blake, Chuck Barnes, and Andrea Sella for sharing crystal growing methods