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Some Tricks for the Single Crystal Growth of Small Molecules

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Basel, Switzerland on the 29th August 2016



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Importance of single crystal growth

- Prerequisite for single crystal X-ray analysis, a fast analytical method that yields the three dimensional arrangement of the elements within the crystal.
- Despite many technical advances, be it on the instrumental (X-ray beam, detector) or on the theoretical side:
 Still **single** crystals needed, ideally with dimensions of about $0.05 \times 0.05 \times 0.2 \text{ mm}^3$
- Crystal polymorphs also play an extremely crucial role in terms of processing, bioavailability, stability, regulatory affairs, and intellectual property protection.^[1]



Techniques for single crystal growth

- In liquid phases: Main goal to achieve supersaturation that is followed by nucleation/crystal growth
- Sublimation sometimes also helpful
- Achievement of supersaturation:
 - cooling (from hot oil bath, in fridge or deep freezer)
 - reduction of solvent amount (evaporation, forgotten NMR tubes...)
 - change of solvent (vapor diffusion / layering)
- From pure liquids at low temperature

P. G. Jones *Chem. Brit.* **1981**, 17, 222;

P. van der Sluis *et al. J. Appl. Crystallogr.* **1989**, 22, 340;

J. Hulliger *Angew. Chem. Int. Ed.* **1994**, 33, 143;

A. J. Blake www.nottingham.ac.uk/~pczajb2/growcrys.htm

G. Santiso-Quiniones, I. Krossing *Z. Anorg. Allg. Chem.* **2008**, 634, 704



Problems for a single crystal analysis

- Chemical purity / identity (e.g. crystals from ^{99m}Tc chemistry)
- Oils / Powders
- Microcrystalline / too small
- Intergrown / twinned
- Not diffracting despite fair size (phase transition?)
- Not solvable
- Not enough data (too weak reflections)
- Residual electron density too high / “unreasonable” electron density

How to overcome the problem of “bad” crystals

- Common situation: crystallographer wants better crystals
 - Despite many references, it is not always clear how to optimize the crystals
 - Often only a few milligrams available!
- ⇒ No systematic study of the solubility

P. G. Jones *Chem. Brit.* **1981**, 17, 222

P. van der Sluis, *et al. J. Appl. Crystallogr.* **1989**, 22, 340

J. Hulliger, *Angew. Chem. Int. Ed.* **1994**, 33, 143

J. Lu and S. Rohani, *Curr. Med. Chem.*, **2009**, 16, 884

A. J. Blake, www.nottingham.ac.uk/~pczajb2/growcrys.htm

P. D. Boyle, www.xray.ncsu.edu/GrowXtal.html



Super Nova with Atlas CCD
Mo/Cu microfocus



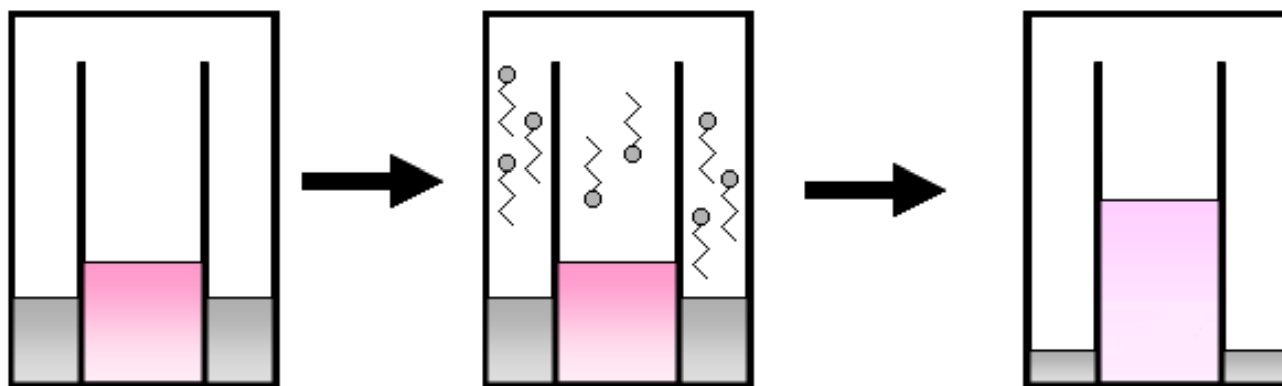
Vapor diffusion

- About 4 mg of substance are dissolved in about 0.5 ml solvent in the inner container.
- About 2.5 ml of antisolvent (normally having a boiling point 5-10 °C **higher** than solvent) are placed in outer container.
- Wait for days, or a few weeks!
- If (anti)solvents have equilibrated and nothing happened, unscrew vial a bit:
 - evaporation experiment from a solvent mixture.

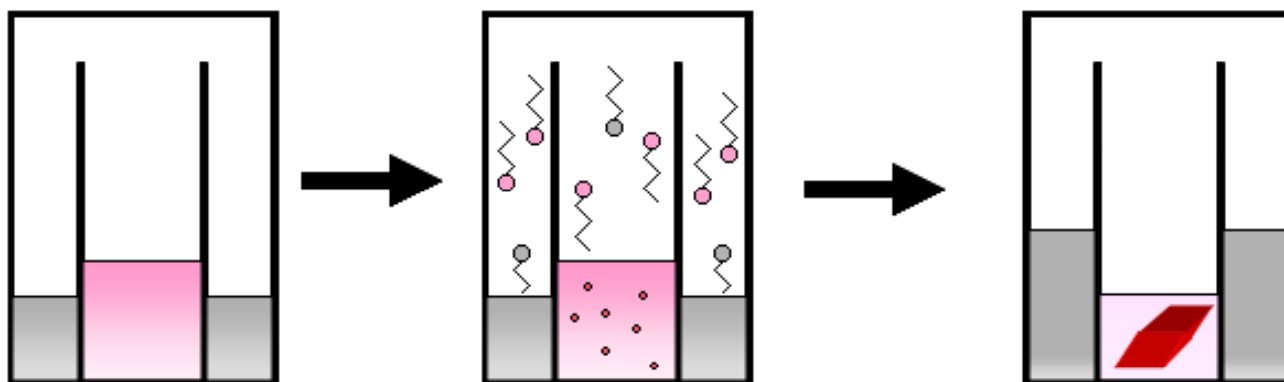


Influence of the boiling points

Antisolvent (e.g. diethylether) has a lower boiling



Antisolvent has a higher boiling point than solvent:





Initial solvent choice for vapor diffusion

Use information gained during synthesis/purification!

Solvent	Antisolvent
tetrahydrofuran	cyclohexane
methylformate	cyclopentane or hexane (dries out)
methylene chloride	cyclopentane
ethanol	cyclohexane
methanol	hexane or tetrahydrofuran
acetonitrile	tetrahydropyran
acetone	chloroform
water	dioxane



Optimization of the vapor diffusion

Improving crystal form/size:

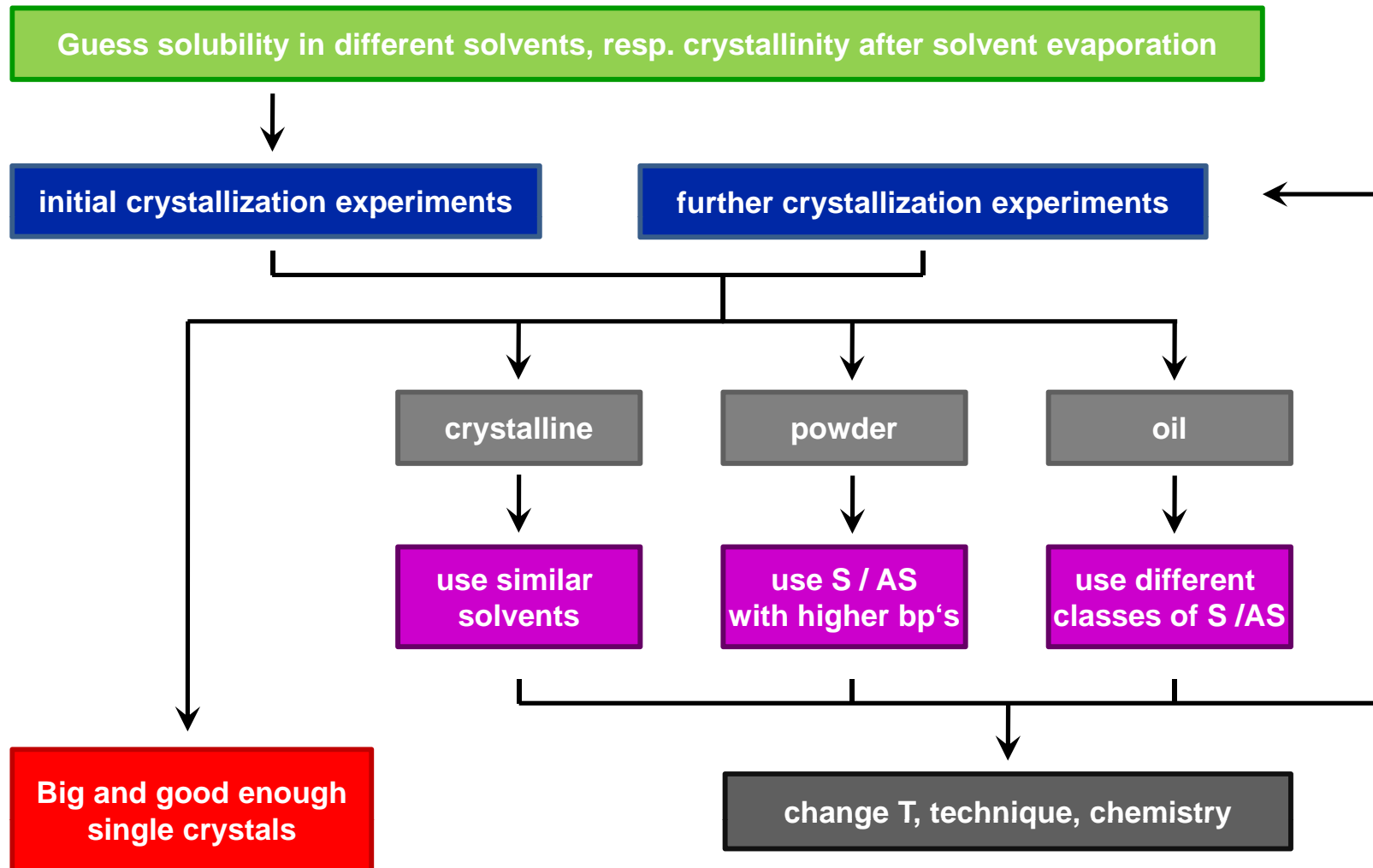
Substitute solvent and antisolvent with other solvents of the same class having similar dielectric constants.

For example:

Solvent	B.p.°C / ϵ	Antisolvent	B.p.°C / ϵ
methylene chloride	40 8.93	cyclopentane	49 1.97
1,1,1-trichloroethane	74 7.24	cyclohexane	81 2.02
1,2-dichloroethane	84 10.4	methylcyclohexane	101 2.02



An extremely simplified flow chart



Layering I

- About 4 mg of substance are dissolved in ~0.5 ml of a dense solvent and put in a cheap NMR tube.
- With an extra-long Pasteur pipette, a 0.5 cm high protection layer of pure solvent is carefully layered above.
- The lighter antisolvent is carefully layered above, until the NMR tube is full.





Layering II

- Rather high boiling solvents are used (e.g. dibutyl ether than diethyl ether).
- Different densities of the solvent and the antisolvent are needed. Take change of density due to solute into account!
- Experiment takes more time to equilibrate (several weeks!).
- Once started the experiment is difficult to modify.
- Difficult optical crystal evaluation, if they grow at the bottom of the NMR tube (or when falling down during retrieval attempts).



Excerpt from a table with 107 solvents

Sum formula	Name	b.p. °C	δ	ϵ
C ₆ H ₁₂ O ₂	t-Butyl acetate	95	0.867	5.67
C ₆ H ₅ Cl	Chlorobenzene	132	1.106	5.69
C ₅ H ₁₀ O ₂	Ethyl propanoate	99	0.892	5.76
C ₄ H ₈ O ₂	Ethyl acetate	77	0.900	6.08
C ₅ H ₁₀ O ₂	Butyl formate	106	0.889	6.10
C ₄ H ₈ O ₂	Methyl propanoate	80	0.915	6.20
C ₂ H ₄ O ₂	Acetic acid	118	1.045	6.20
C ₄ H ₈ O ₂	Propyl formate	81	0.906	6.92
C ₅ H ₁₀ O	2-Methyltetrahydrofuran	78	0.855	6.97
C ₃ H ₆ O ₂	Methyl acetate	57	0.934	7.07
C ₆ H ₁₄ O ₃	Diethylene glycol dimethyl ether	162	0.943	7.23
C ₂ H ₃ Cl ₃	1,1,1-Trichloroethane	74	1.339	7.24
C ₄ H ₁₀ O ₂	Ethylene glycol dimethyl ether	85	0.869	7.30
C ₄ H ₈ O	Tetrahydrofurane	65	0.889	7.52
CH ₂ Br ₂	Dibromomethane	97	2.497	7.77
C ₂ H ₂ Cl ₈	1,1,2,2-Tetrachloroethane	131	1.541	8.50
C ₃ H ₆ O ₂	Ethyl formate	54	0.917	8.57

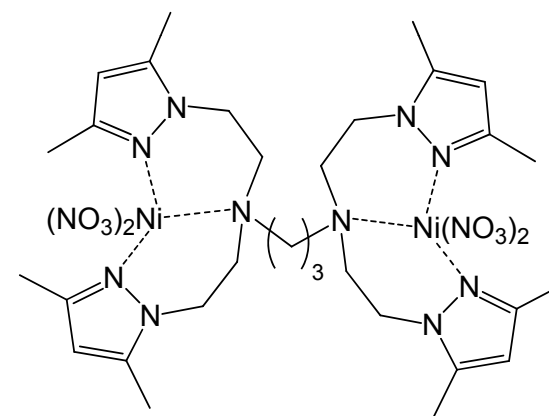
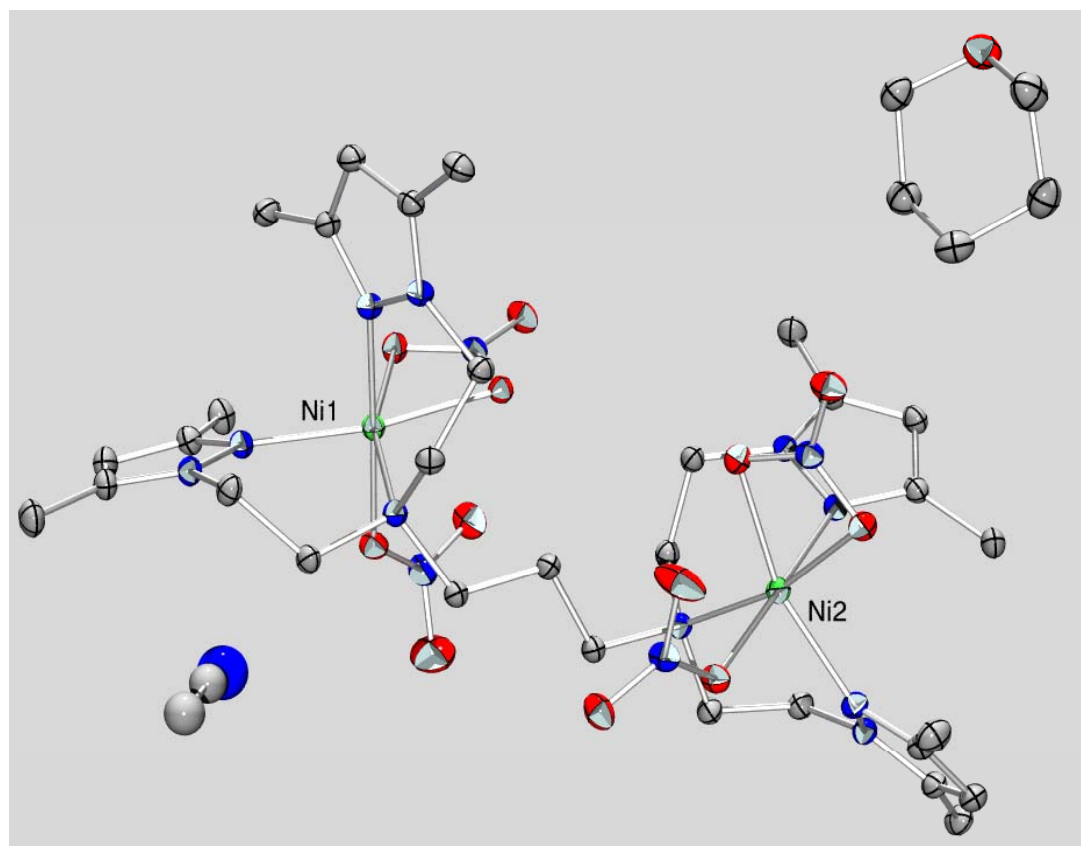
selection criteria for these solvents:

- m.p. < 20° C
- b.p. > 30° C
- few with b.p. > 150° C
- stability
- toxicity
- cost

Example I

Recrystallization from acetonitrile versus tetrahydropyran

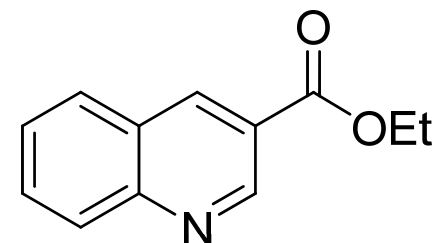
Single crystals from methanol versus tetrahydropyran



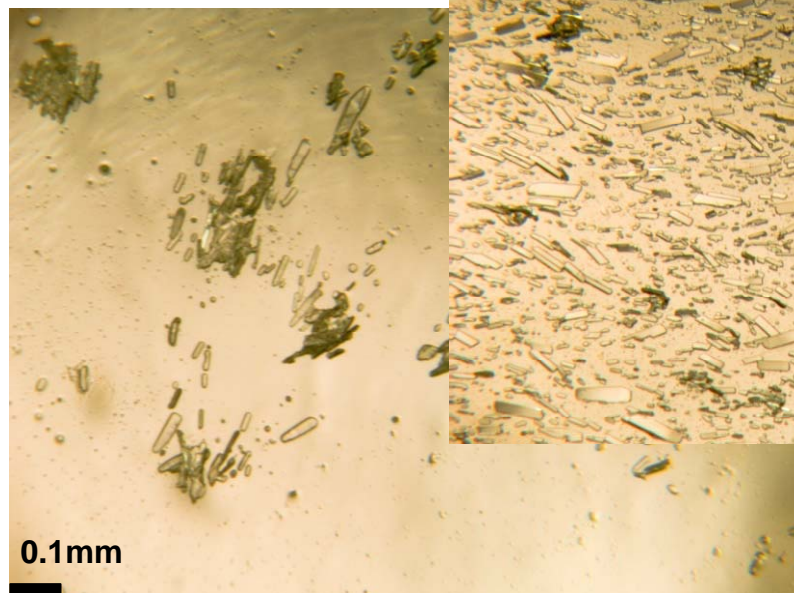
B. Spingler, P. M. Antoni
Chem. Eur. J. **2007**, *13*, 6617

Example II

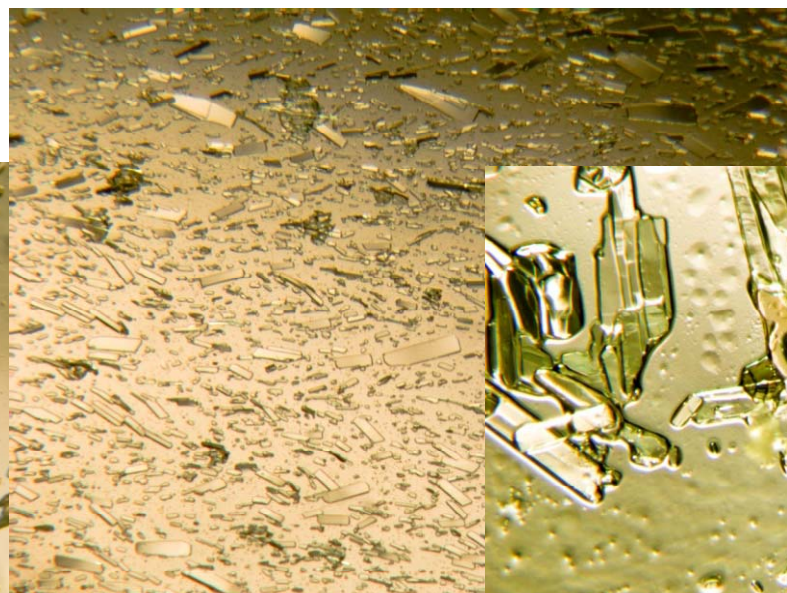
3-carbethoxyquinoline (vapor diffusion):



THF versus
cyclohexane



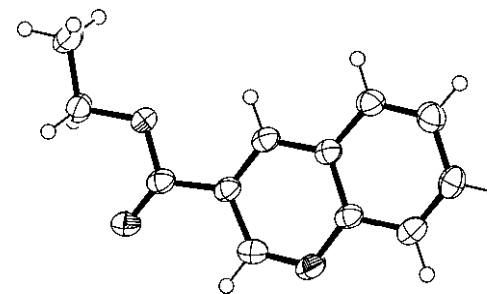
Chloroform versus cyclohexane



Trichloroethylene versus heptane



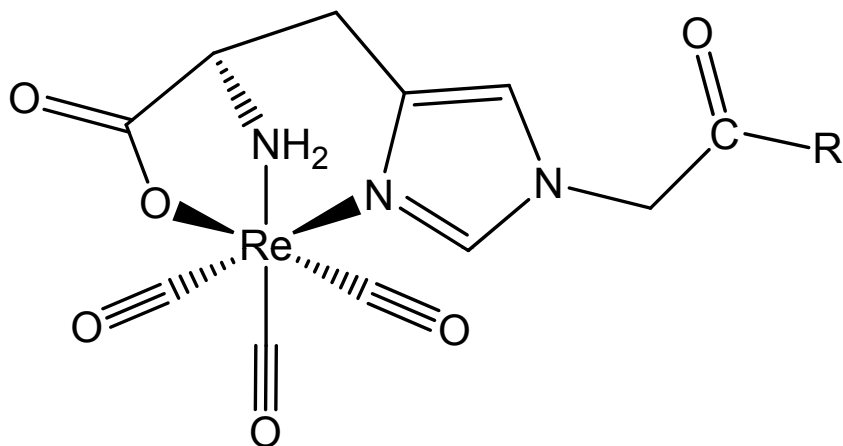
Example II



Formula	C ₁₂ H ₁₁ NO ₂
Space group	P-1
a [Å]	7.5975(5)
b [Å]	12.2026(7)
c [Å]	12.8137(8)
α [°]	61.607(6)
β [°]	77.247(5)
γ [°]	78.353(5)
Volume [Å ³]	1012.74(11)
Z	4

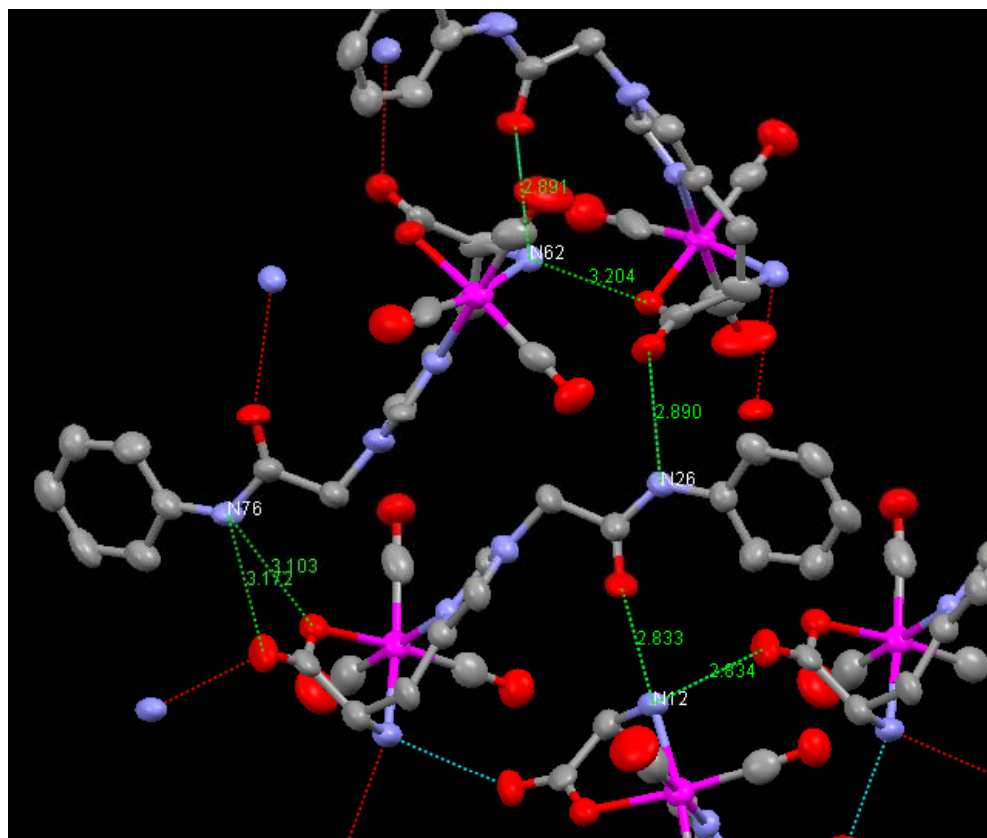
Crystal size [mm ³]	0.21 x 0.13 x 0.08
Wavelength [Å]	0.71073
Independent reflections	5457 [R _(int) = 0.0402]
Reflections observed (>2σ(I))	2329
Completeness to theta	99.9 % to 29.13°
Max. and min. transmission	0.9928 and 0.8558
Data / restraints / parameters	5457 / 0 / 273
Goodness-of-fit on F ²	0.810
Final R indices (I>2σ(I))	R ₁ = 0.0479 , wR ₂ = 0.0782
Largest diff. peak and hole [e.Å ⁻³]	0.163 and -0.268

Example III, different chemistry



R: OH, OEt, Ot-Bu
all did not crystallize.

But R: NHPH did.





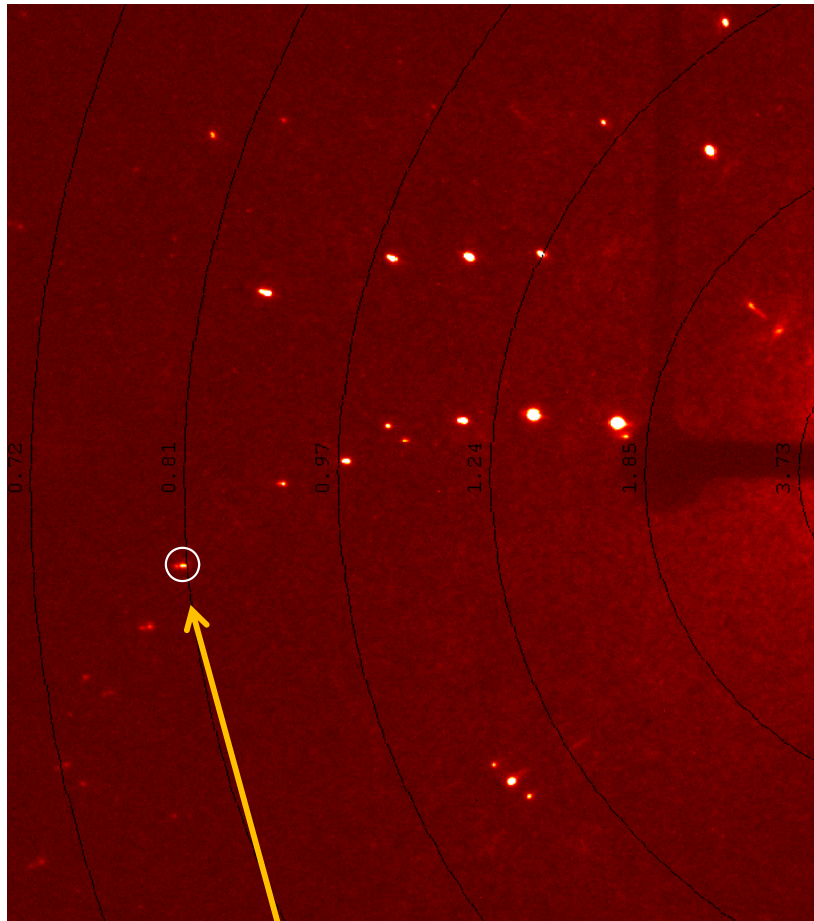
Mo versus Cu radiation?

- Traditional knowledge [1]:
 - Mo for crystals with heavy elements
 - Cu for organic crystals (absorption challenges with heavy elements)
- However with new diffractometer and software systems, Cu became an important rescue option for weakly diffracting crystals of high quality containing only a few heavy elements and mainly light elements

[1] A. J. Blake, J. M. Cole, J. S. O. Evans, P. Main, S. Parsons, D. J. Watkin *Crystal Structure Analysis, Principles and Practice*; Oxford University Press, **2009**, pp. 352

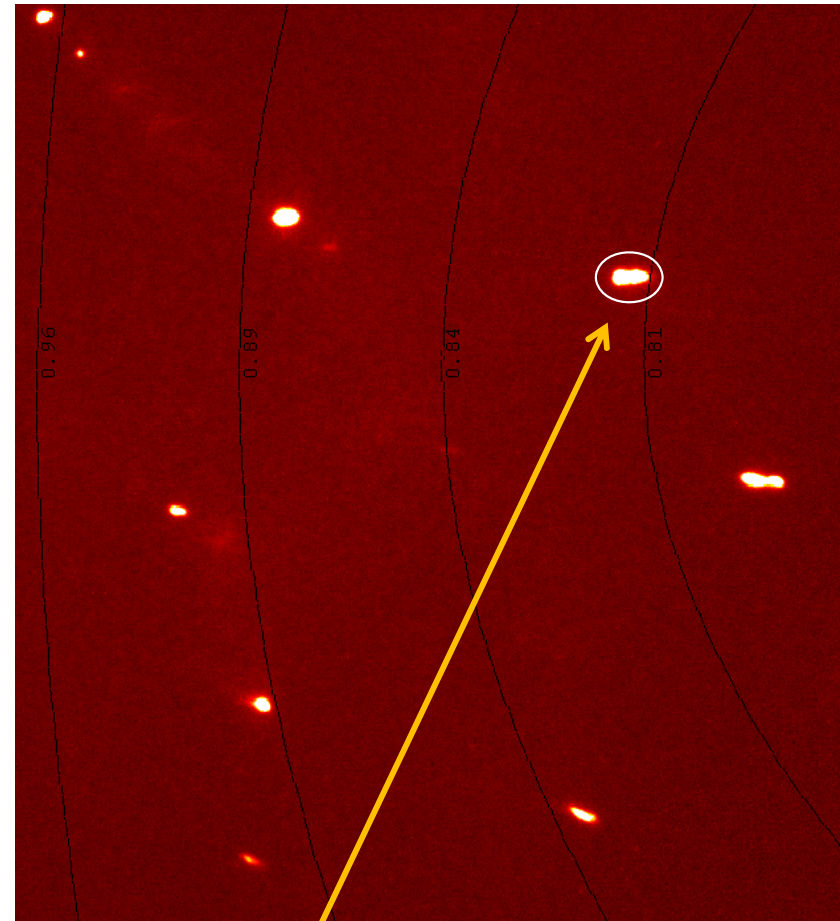


Mo: 60 s exposure



hkl: 1 -10 -21; I/ σ : 13

Cu: 16 s exposure



hkl: 1 -10 -21; I/ σ : 105



Two examples of heavy metal containing structure measured with Cu radiation

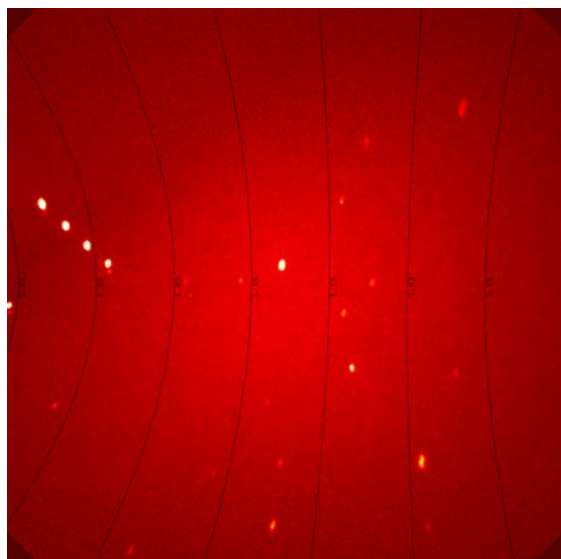
Empirical formula	$C_{39}H_{33.25}Cl_2F_6N_8O_{7.13}PRu$ [1]	$C_{44}H_{58}Br_2CoN_6O_8$ [2]
Diffractometer	SuperNova dual radiation CCD	SuperNova dual radiation CCD
Space group	<i>P</i> -1	<i>P</i> -1
Abs. coeff. (mm ⁻¹)	5.465	5.664
Crystal size (mm ³)	0.33 x 0.08 x 0.04	0.18 x 0.04 x 0.02
Indep. reflections	15295 [$R_{int} = 0.0338$]	4532 [$R_{int} = 0.0218$]
Completeness to θ	95.0 % to 66.97°	99.4 % to 66.97°
Absorption corr.	Semi-empirical from equiv.	Gaussian + Semi-empirical from equiv.
Max. and min. transm.	0.8110 and 0.6223	0.918 and 0.617
Fin. R ind. [$I > 2 \sigma(I)$]	$R1 = 0.0760$, $wR2 = 0.2128$	$R1 = 0.0235$, $wR2 = 0.0586$
Fin. diff. ρ_{max} (e-/Å ⁻³)	1.356 and -1.533	0.544 and -0.324

[1] C. Mari, *et al. Chem. Eur. J.* **2014**, *20*, 14421

[2] E. Joliat, *et al. Dalton Trans.* **2016**, *45*, 1737

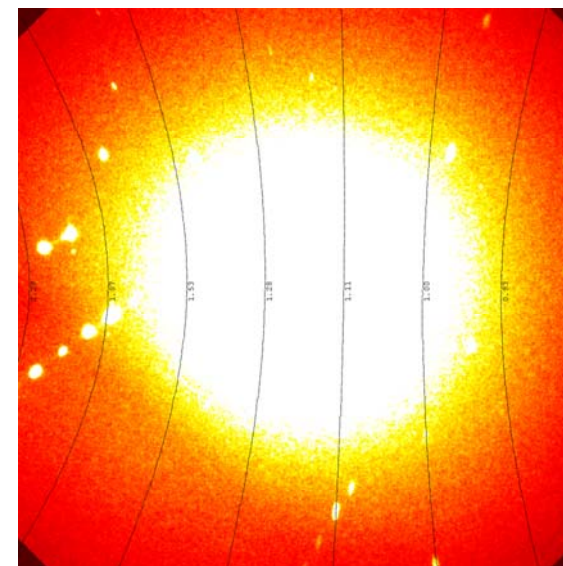
Disadvantages of Cu radiation

- Only till 0.8 Å resolution
- Big theta values to be covered mean 3 series of scans (→ longer measurement times)
- For cobalt containing compounds and weakly diffracting crystals: observation of X-ray fluorescence



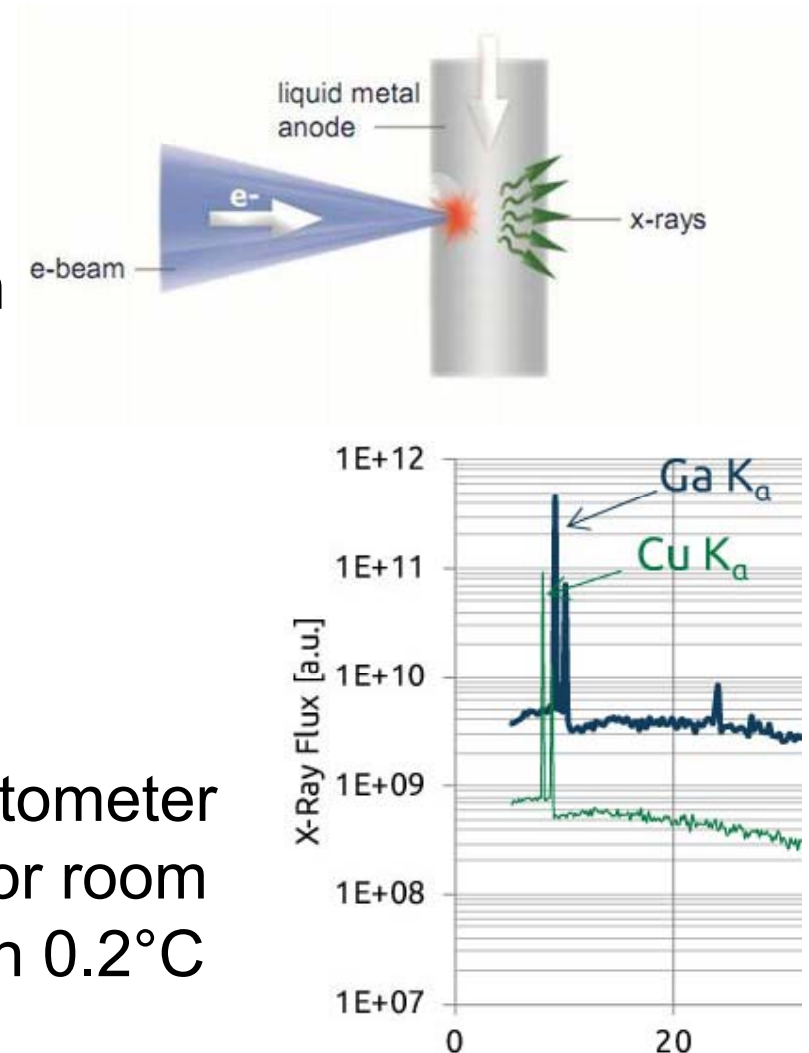
Left: 6s exposure
2.55-0.95 Å

Right: 24s exposure
2.29-0.93 Å



What about Ga radiation?

- Melts at 29.8°C, but actually Ga rich alloys being used
- Liquid metal dissipates heat much quicker than solid one
- Most intensive microsource
- Wavelength 1.34 Å
- No cobalt fluorescence observed
- Air-conditioning needed for diffractometer room, as Ga source most stable for room temperature not varying more than 0.2°C

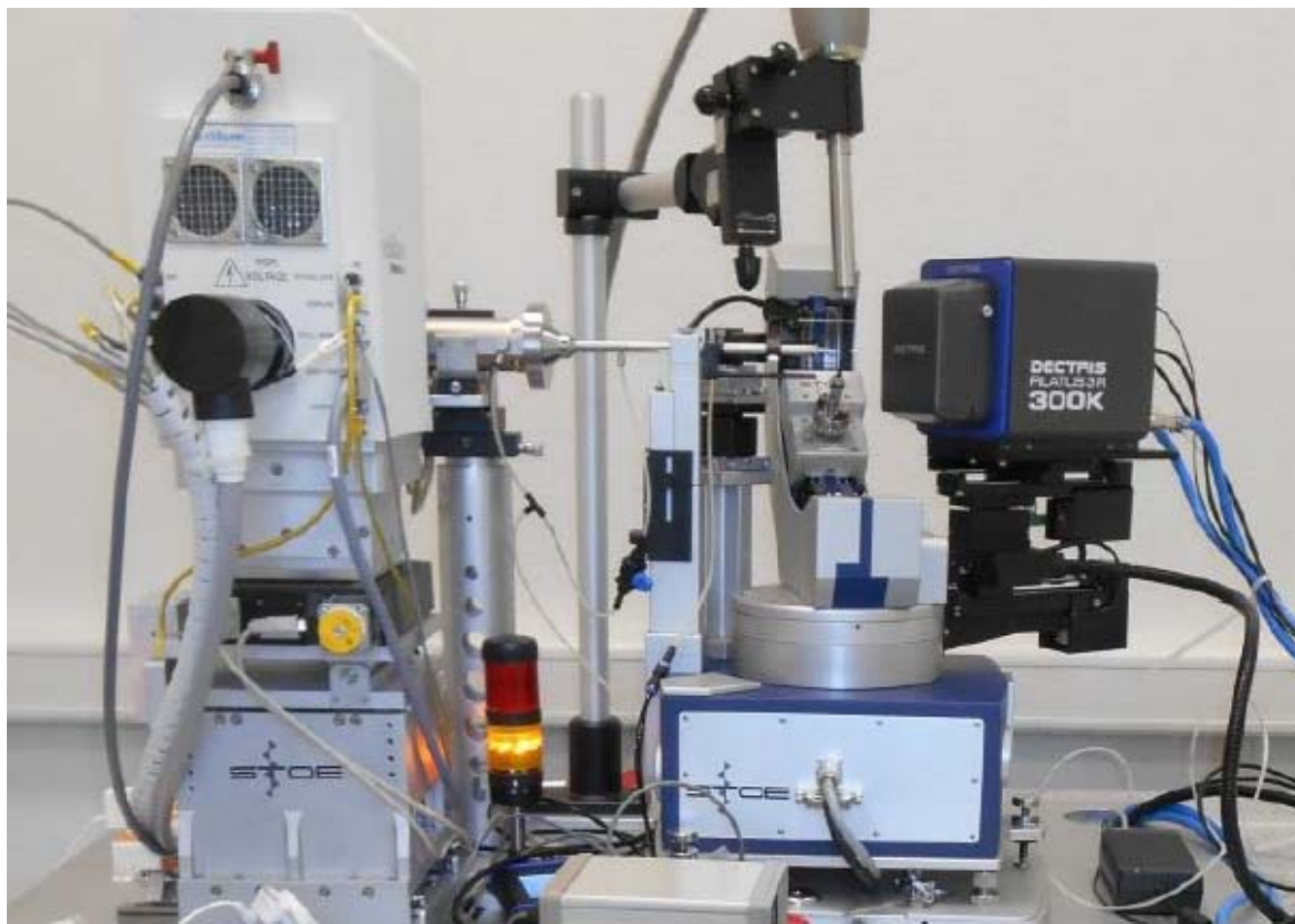


[1] Pictures taken from www.excillum.com/technology/metal-jet-technology.html

[2] M. Otendal, T. Tuohimaa, U. Vogt, H. M. Hertz *Rev. Sci. Inst.* **2008**, 79, 016102-3



Ga jet with a Pixel detector



Setup at the
University of
Basel,
Switzerland

See: L. Prieto *et al.* *Org. Lett.* **2016**, *18*, 5292



Summary

- Stay with your system as long as it works!
- For optimization of unsatisfactory crystals:
 - Systematically explore the crystallization properties of a solvent class with a similar dielectric constant
 - use an anti-/solvent pair with an inversed polarity
 - change the technique
 - change the anions, add additives
- Do not be afraid of copper radiation, even if you have some heavy elements present!



Acknowledgements

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