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# **Special cases**

**Anomalous dispersion**

**Extinction**

**Violation of systematic absences**

**If things went wrong...**

- **Absolute Configuration** : spatial arrangement of the atoms for a chiral molecule ( $R/S$ ,  $P/M$ ,  $\Delta/\Lambda$ ,  $(+)/(-)$  or  $D/L$  assignment).
- **Absolute Structure** : spatial arrangement of atoms in a non-centrosymmetric crystal structure (unit-cell, space group)
- **Polar space group** : Non-centrosymmetric space group containing mirror plans. E. g. :  $Pc$ ,  $Pna2_1$
- **Chiral space groups / Sohncke space groups** : Space groups containing no inversion center, mirror or glide planes. E. g.  $P1$ ,  $P2_1$ ,  $P3_1$ ,  $P4_1$
- **Enantiomorph**: The mirror image of a chiral crystal
- **Enantiomorphic space group**: A space group whose mirror image is not the same space group. E. g.  $P4_1|P4_3$ , but not  $P1$  or  $P2_1$

# In the solid state

- Chiral molecules can crystallize as an enantiopure bulk sample or as a **racemic mixture**.
- Enantiopure compounds/enantiopure crystals → Space group restriction:
  - Only 65 space groups allowed (chiral or Sohncke space groups)
  - No inversion centers / no mirror planes / no glide planes

Triclinic:  $P1$

Monoclinic:  $P2$   $P2_1$   $C2$

Orthorhombic:  $P222$   $P222_1$   $P2_12_12$   $P2_12_12_1$   $C222_1$   $C222$   $F222$   $I222$   $I2_12_12_1$

Tetragonal:  $P4$   $P4_1$   $P4_3$   $P4_2$   $I4$   $I4_1$   $P422$   $P42_12$   $P4_122$  |  $P4_322$   $P4_12_12$  |  $P4_32_12$   $P4_222$   $P4_22_12$   $I422$   $I4_122$

Trigonal:  $P3$   $P3_1$  |  $P3_2$   $R3$   $P3_12$   $P321$   $P3_112$  |  $P3_212$   $P3_121$  |  $P3_221$   $R32$

Hexagonal:  $P6$   $P6_1$  |  $P6_5$   $P6_2$  |  $P6_4$   $P6_3$   $P622$   $P6_122$  |  $P6_522$   $P6_222$  |  $P6_422$   $P6_322$

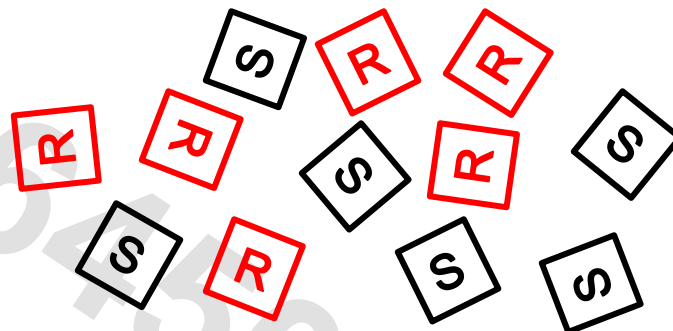
Cubic:  $P23$   $F23$   $I23$   $P2_13$   $I2_13$   $P432$   $P4_232$   $F432$   $F4_132$   $I432$   $P4_132$  |  $P4_332$   $I4_132$

- These include 11 pairs of **enantiomorphic space groups** (screw axes of opposite handedness). For these, inversion of the crystal generates another space group.

# Racemic mixtures in the solid state

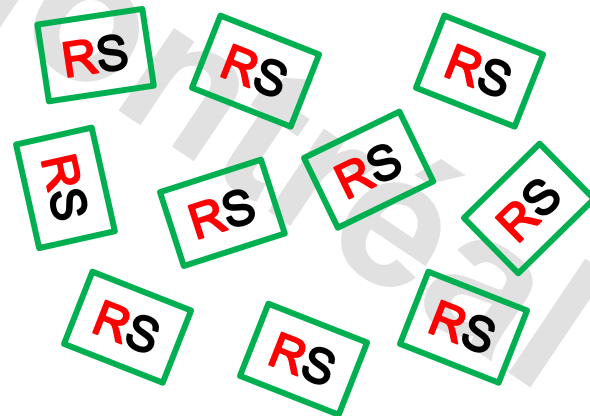
1) **Conglomerate**: a mixture of well-resolved crystals of both enantiomers

- Chiral space group
- Individual crystals are enantiopure and have optical activity



2) **Racemate**: Each crystal contains both enantiomers in equal amounts.

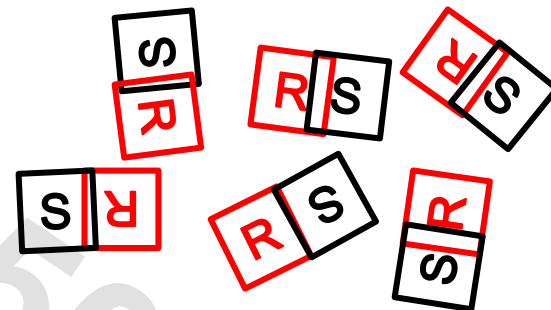
- No optical activity
- All space groups possible
- Typically centrosymmetric space group



# Racemic mixtures in the solid state

## 3) **Inversion twin**: twinned crystals of both enantiomers

- Chiral space group
- Ratio of enantiomers not necessarily 1:1

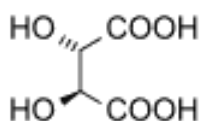
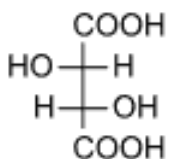
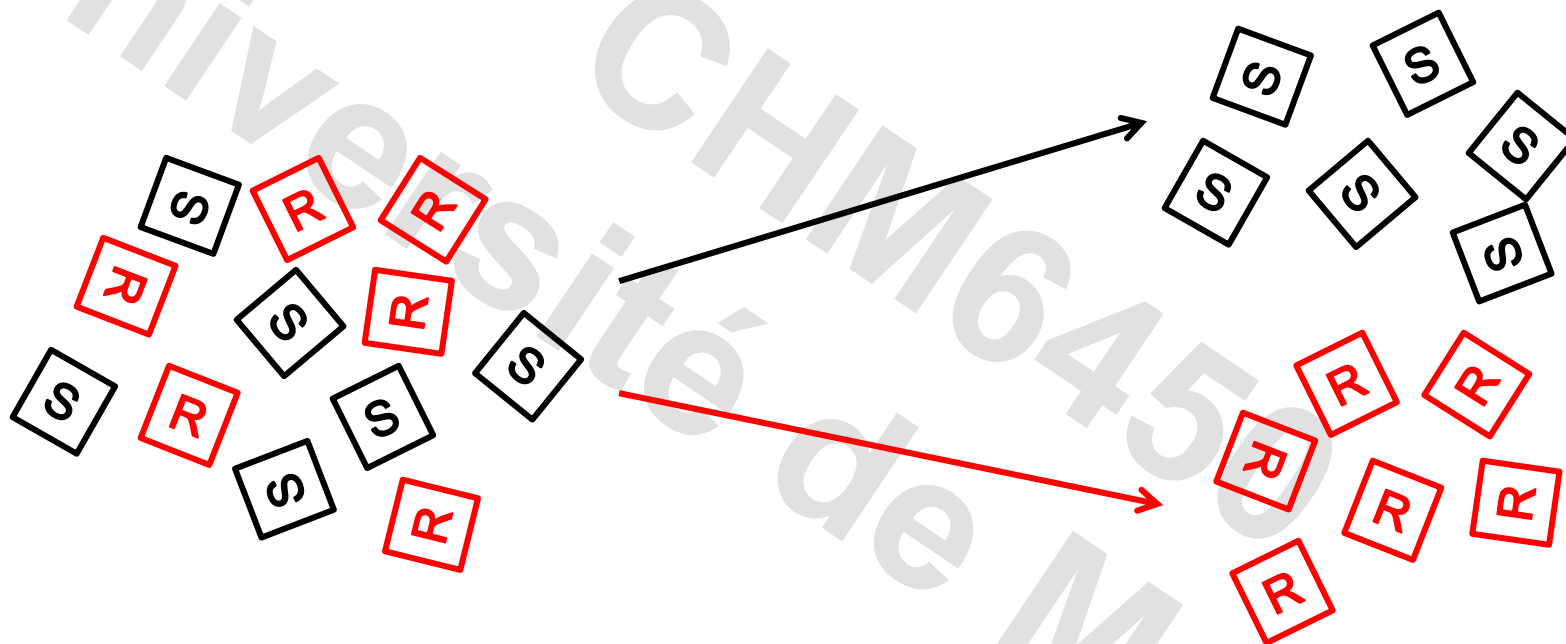


## 4) **Disordered solid-solution**: Crystal containing the two enantiomers in a disordered arrangement.

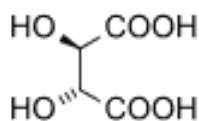
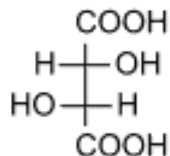
- Usually centrosymmetric space group



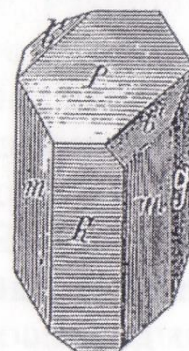
# Louis Pasteur (1848): Separation of the two enantiomers by the visual sorting of crystals of a conglomerate



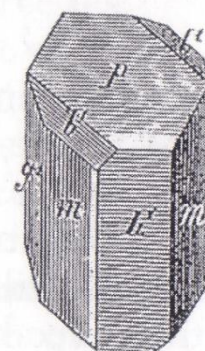
D-(-) levotartaric acid



L-(+) dextrotartaric acid

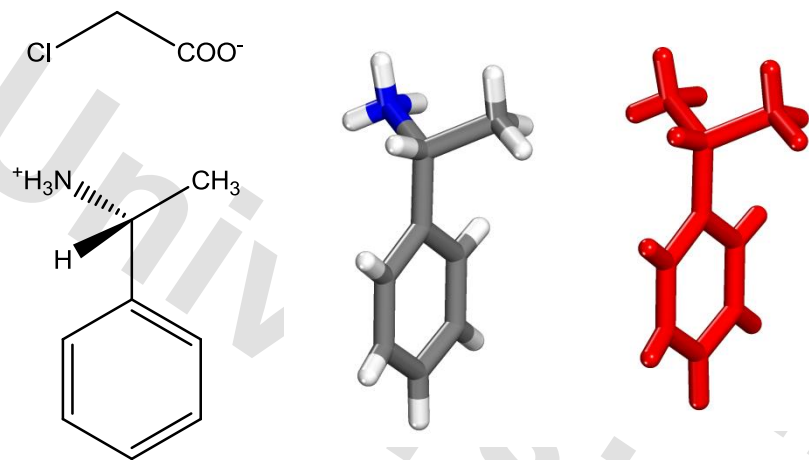


(Droit.)

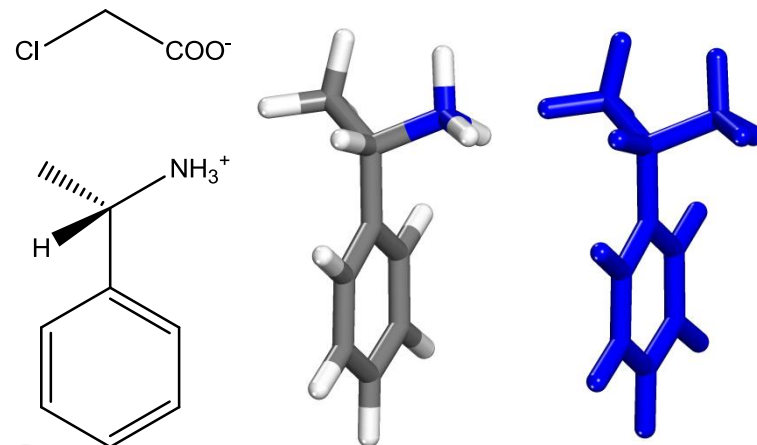


(Gauche.)

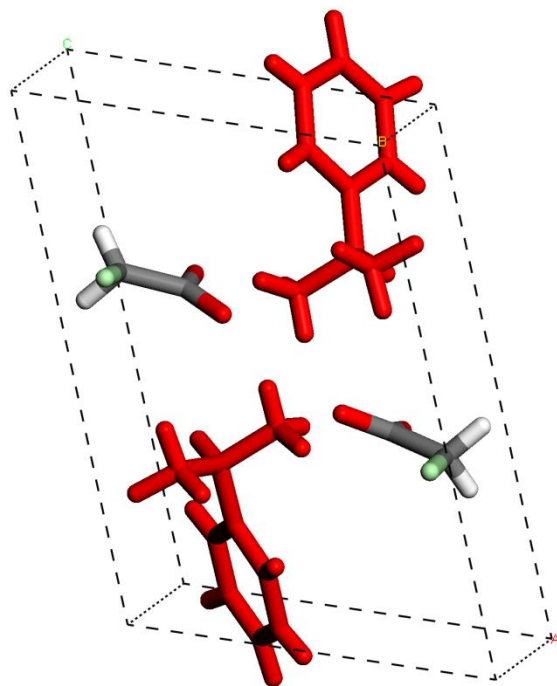
Sodium ammonium tartrate crystals



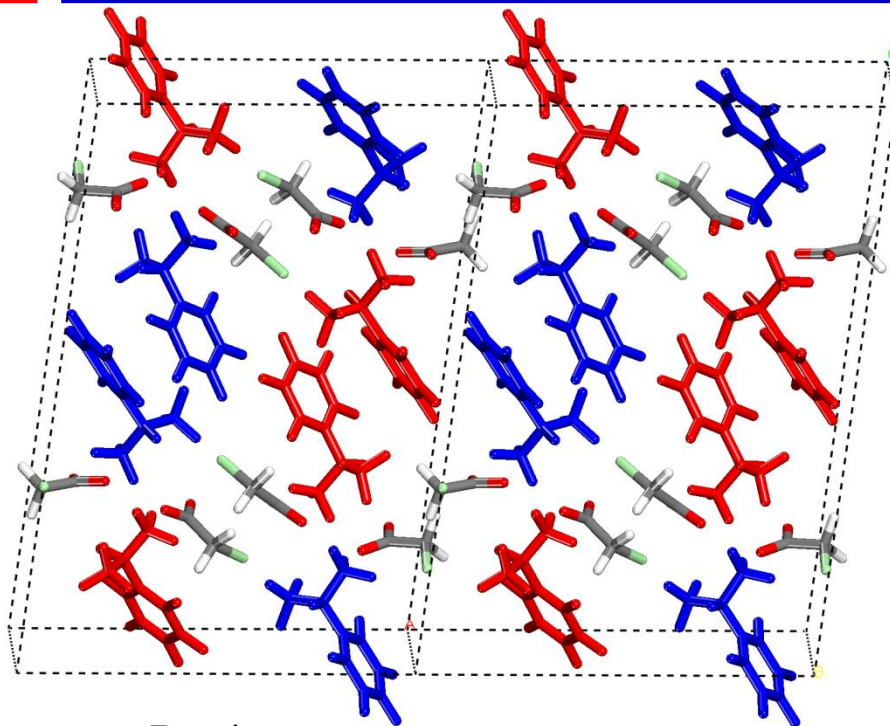
(*R*)-1-phenylethanaminium



(*S*)-1-phenylethanaminium

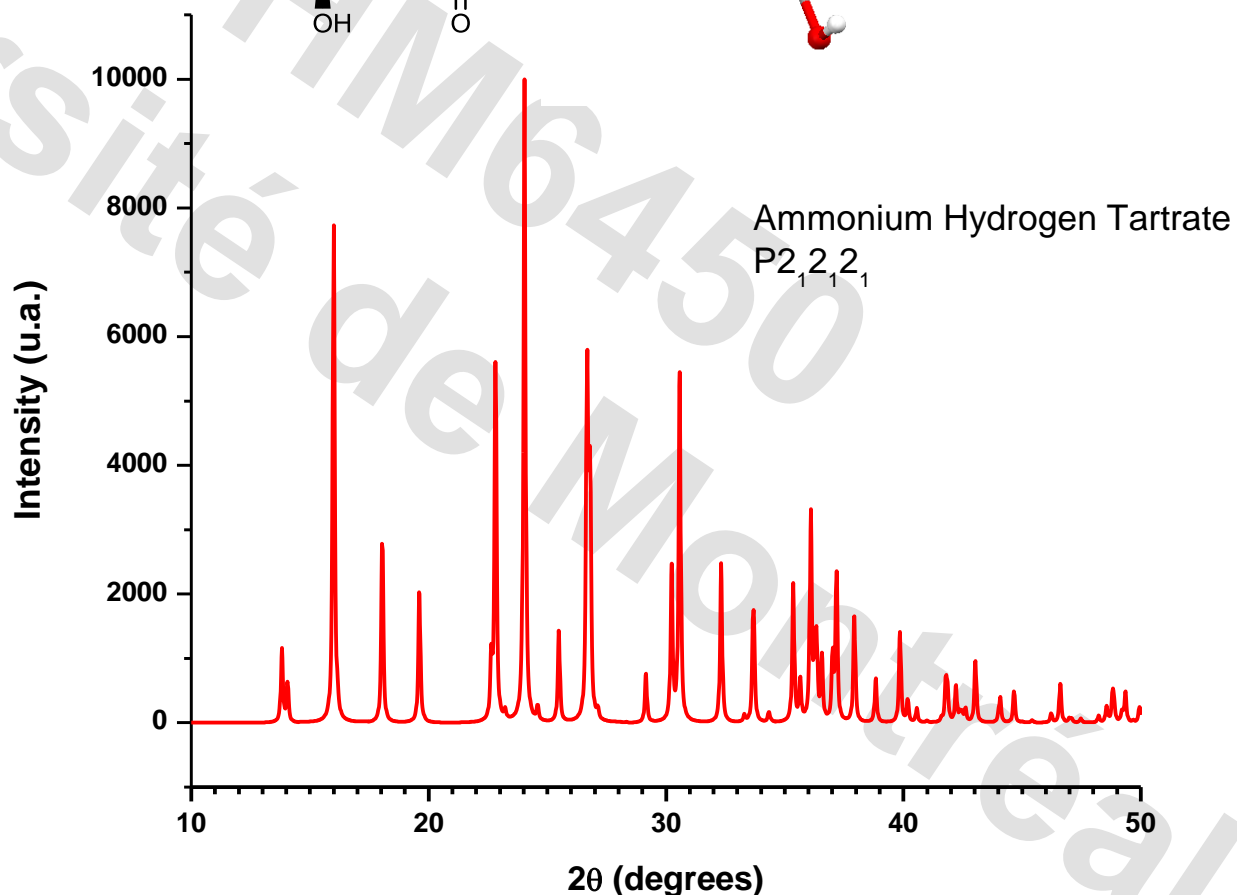
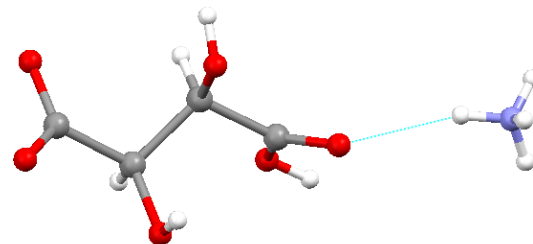
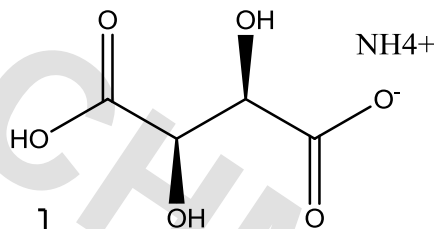
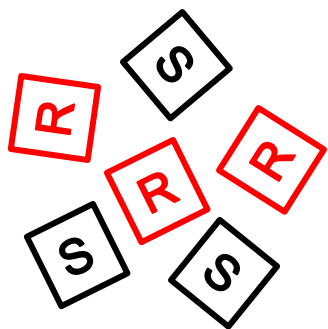
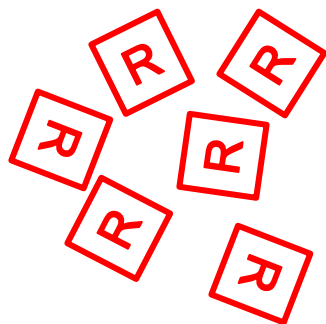
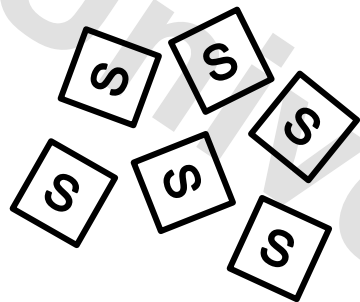


$P2_1$



$P2_1/c$

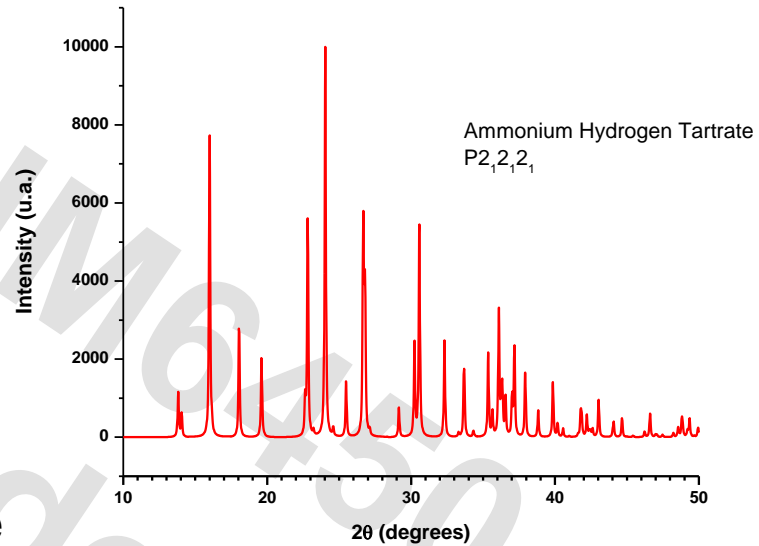
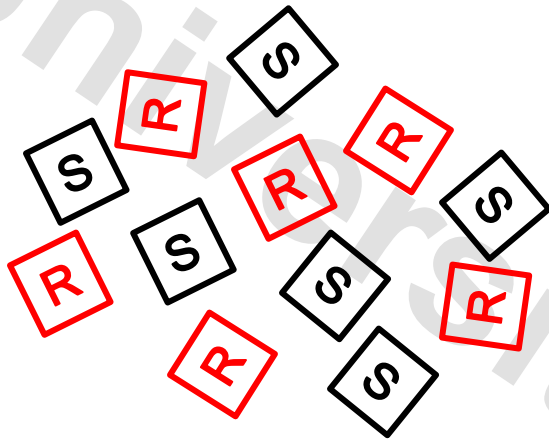
# Powder diffraction



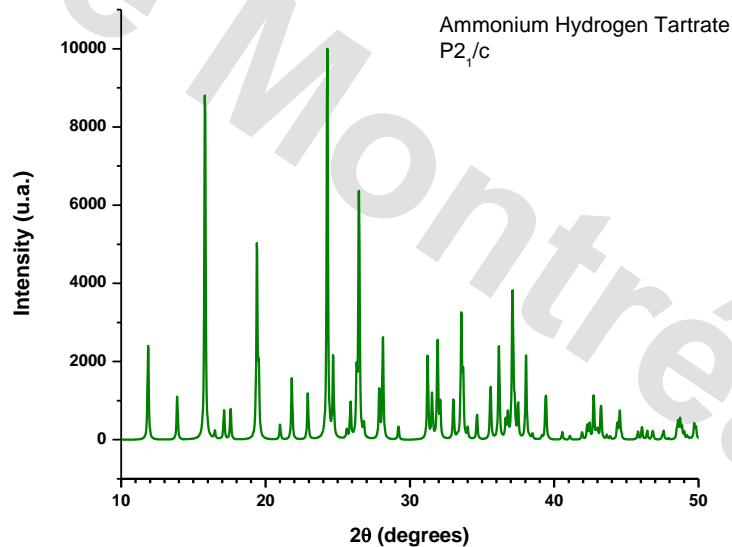
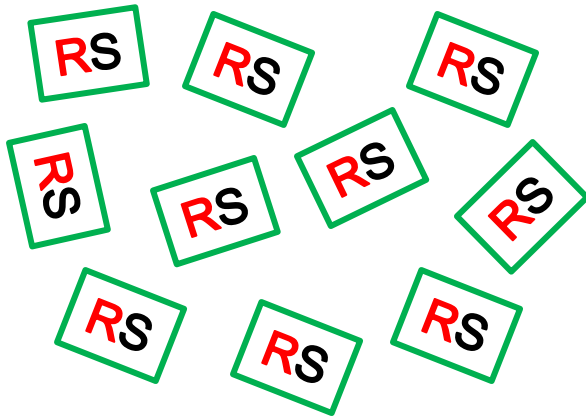
The two enantiopure samples and the conglomerate give the same powder X-ray diffraction pattern



# Powder diffraction

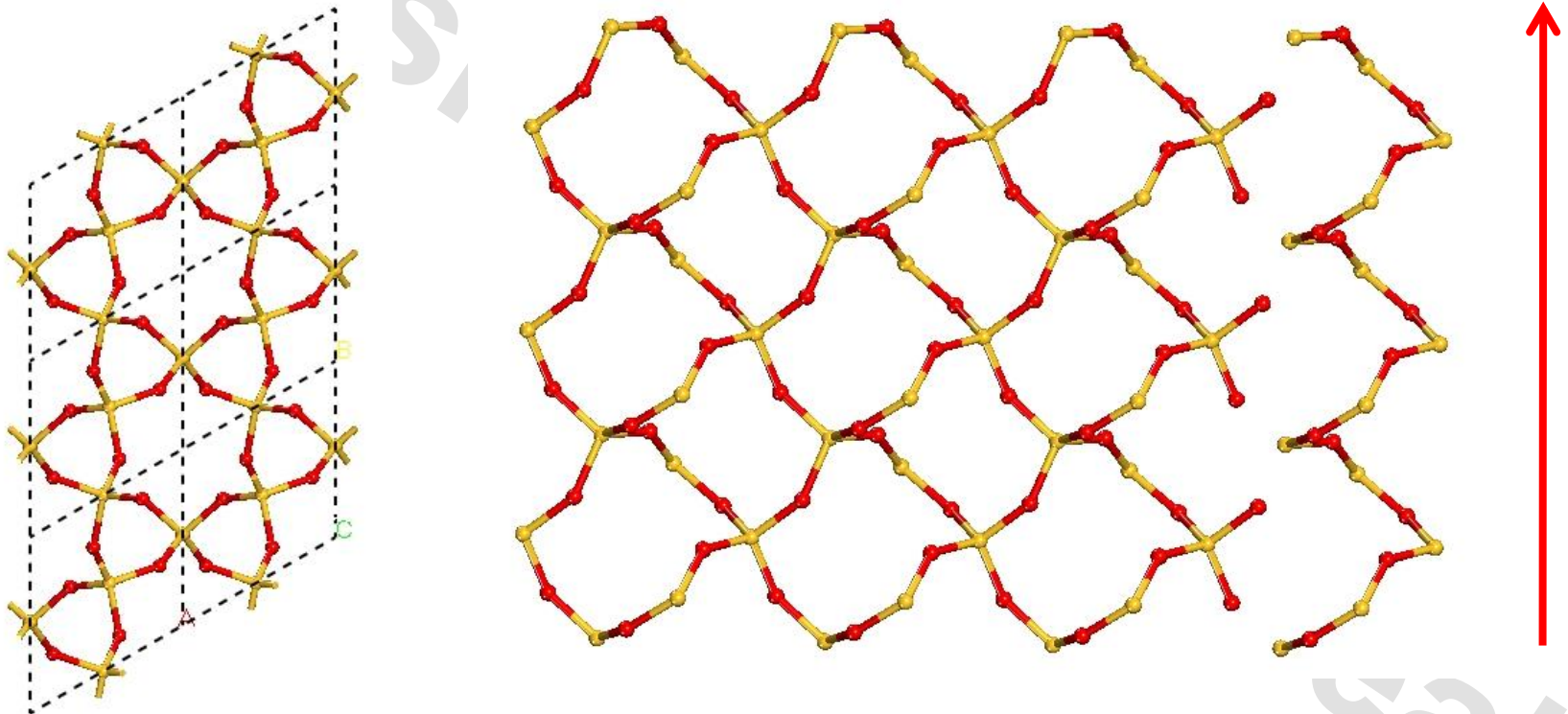


But the conglomerate and racemate have different space groups and powder diffraction patterns



# Polar structures

Achiral molecules can crystallize in chiral and non-centrosymmetric space groups. While the *molecule* is *not* chiral, their spatial arrangement in the crystal *is chiral*.



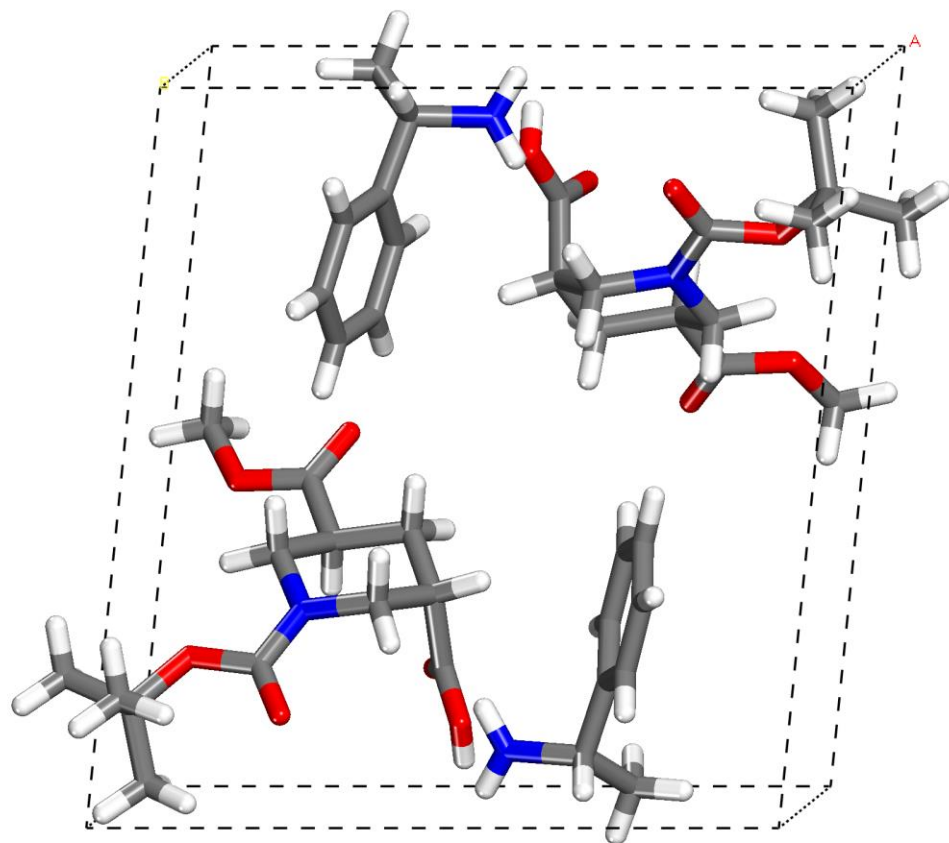
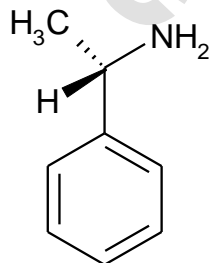
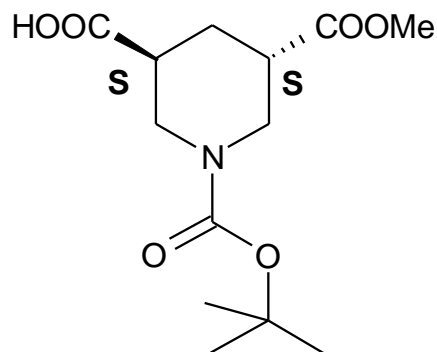
Helical arrangement of  $\text{SiO}_4$  tetrahedra in  $\alpha$ -quartz ( $P3_121$ )

# How do we determine the absolute configuration ?

- Method 1 : Internal chiral reference
- Method 2 : From absolute structure (anomalous dispersion)

# Absolute configuration from an internal chiral reference

Co-crystallisation with a compound of known  
absolute configuration



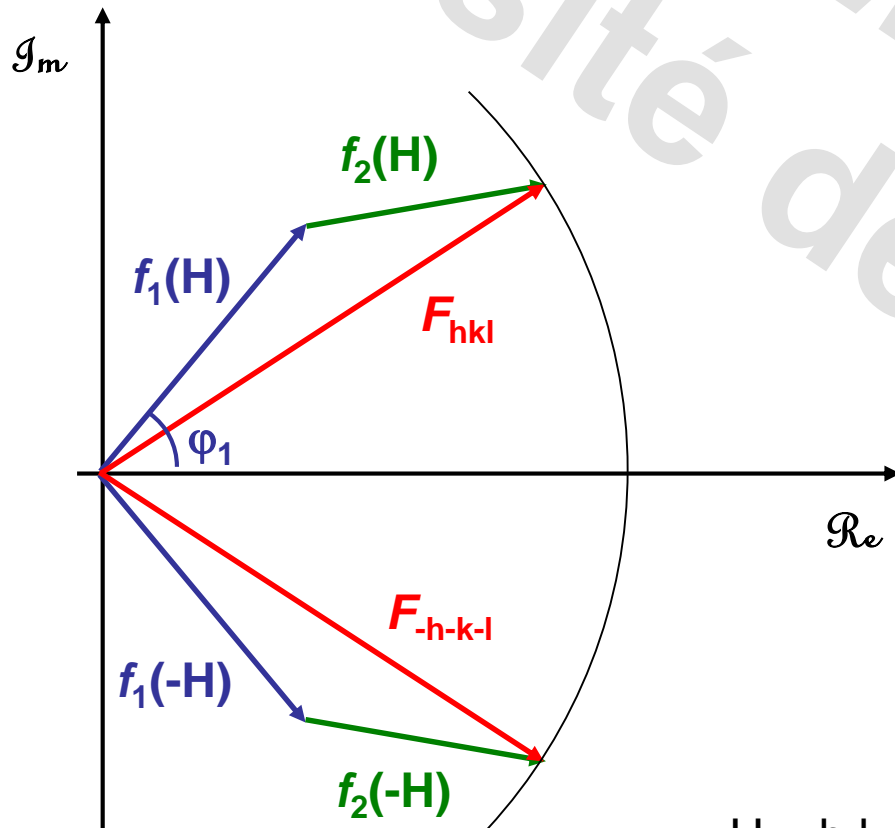
Iding *et al.*,  
Tetrahedron Asymmetry 14 (2003) 1541-1545

# Anomalous dispersion

Again: **Friedel law**

$$F_{hkl} = \sum_{j=1}^N f_j \cdot e^{2\pi i(h \cdot x_j + k \cdot y_j + l \cdot z_j)} = \sum_{j=1}^N f_j \cdot (\cos \varphi_j + i \sin \varphi_j); \quad \varphi_j = 2\pi(h \cdot x_j + k \cdot y_j + l \cdot z_j)$$

$$F_{\bar{h}\bar{k}\bar{l}} = \sum_{j=1}^N f_j \cdot (\cos(-\varphi_j) + i \sin(-\varphi_j)) = \sum_{j=1}^N f_j \cdot (\cos \varphi_j - i \sin \varphi_j)$$



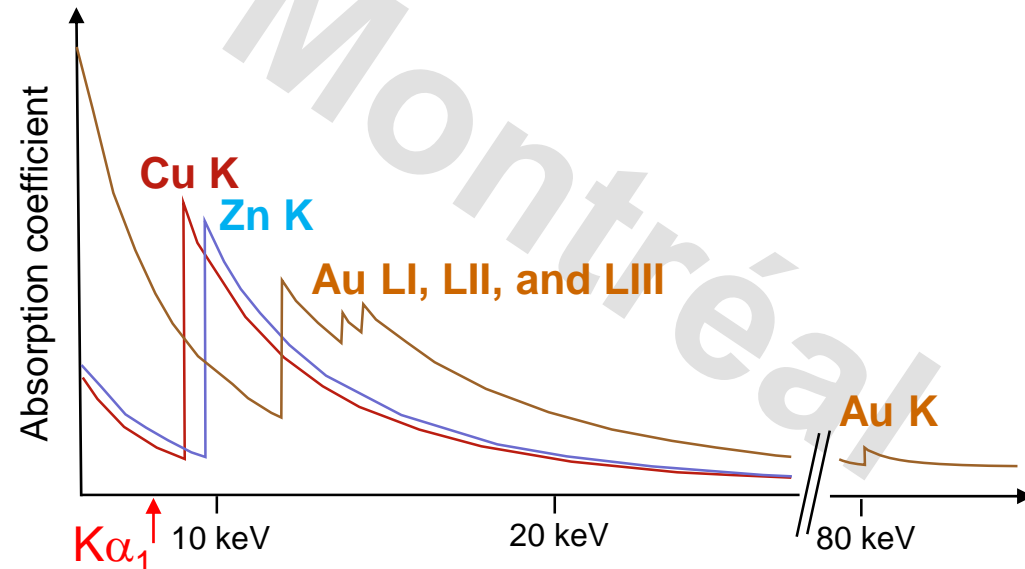
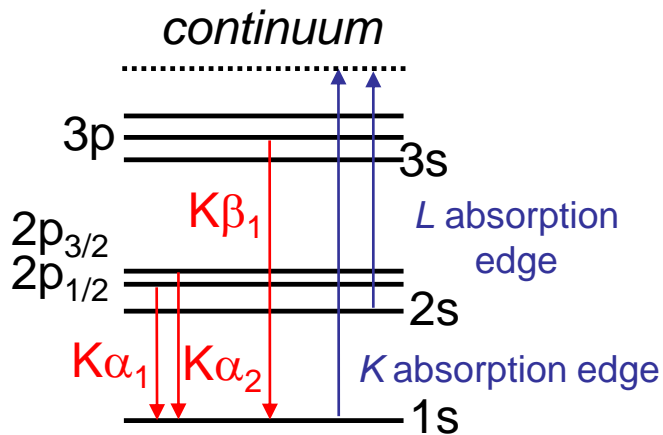
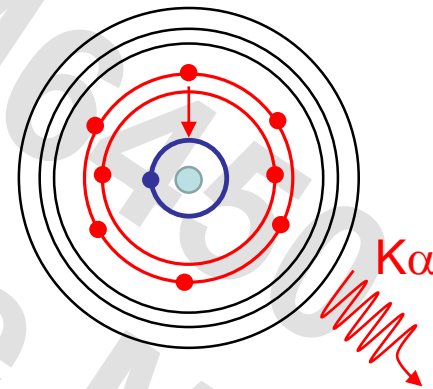
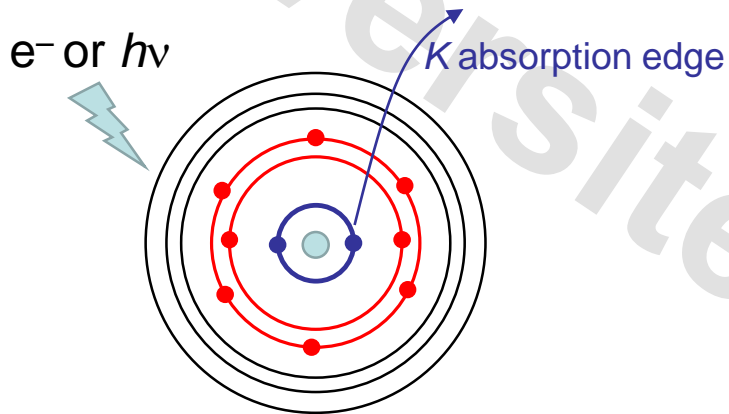
$$|F_{hkl}|^2 = |F_{\bar{h}\bar{k}\bar{l}}|^2$$

$$H = h, k, l; -H = -h, -k, -l$$

# Anomalous dispersion – non-centrosymmetric

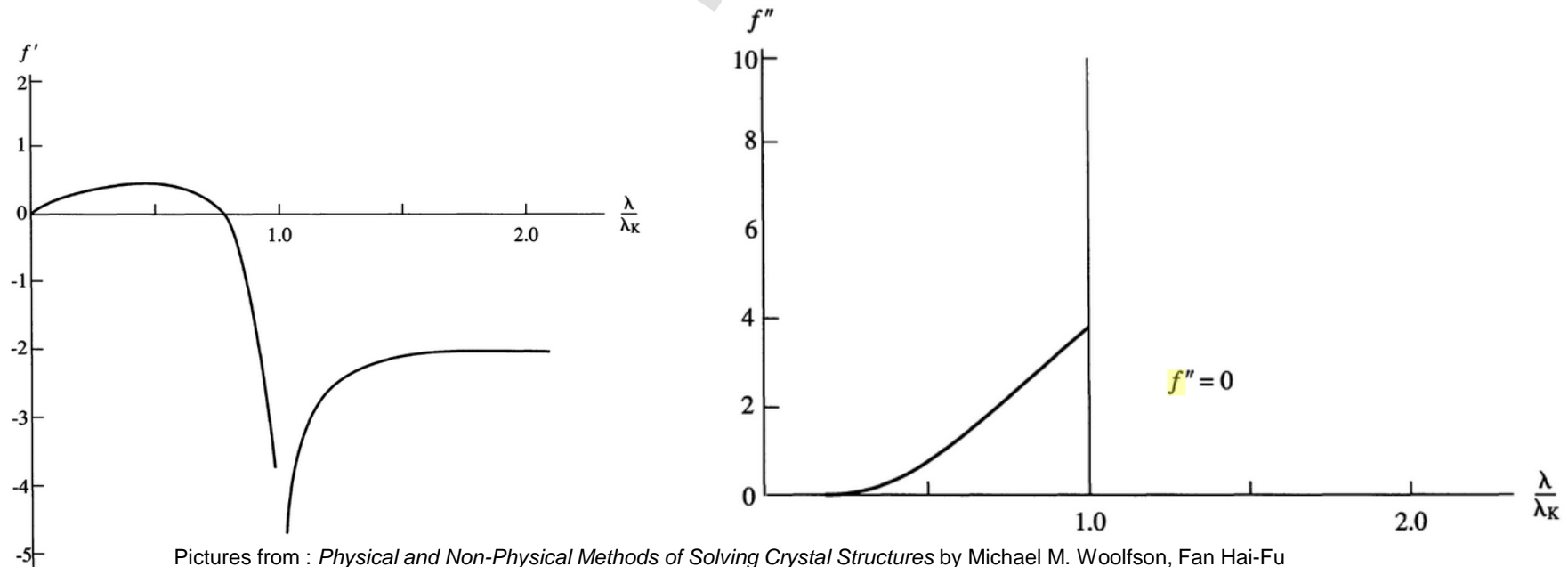
- If the wavelength of the X-rays is **close to an excitation energy of an atom**, we observe a resonance effect which influences the amplitude and the phase of our diffracted photon.

## X-ray absorption and emission:



# Anomalous dispersion – non-centrosymmetric

- If the wavelength of the X-rays is **close to an excitation energy of an atom**, we observe a resonance effect which influences the amplitude and the phase of our diffracted photon.
- We can describe this effect with two correction factors:  $f = f_{\text{atomique}} + \Delta f' + i \cdot \Delta f''$ .
- The real part of the anomalous scattering factor,  $\Delta f'$ , can be positive (0 ° phase shift) or negative (180° phase shift). Close to an absorption edge,  $\Delta f'$  becomes negative and can be related to X-ray absorption.
- The imaginary part of the anomalous scattering factor,  $\Delta f''$ , has a phase shift of 90° and is always positive, large close to the absorption edge and zero at energies lower than the absorption edge.



# Anomalous dispersion – non-centrosymmetric

- If the wavelength of the X-rays is **close to an excitation energy of an atom**, we observe a resonance effect which influences the amplitude and the phase of our diffracted photon.
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- The imaginary part of the anomalous scattering factor,  $\Delta f''$ , has a phase shift of 90° and is always positive, large close to the absorption edge and zero at energies lower than the absorption edge.

## Scattering factors for Cu K $\alpha$ radiation

Element	C	Si	V	Fe	Co	Ni	Cu	Zn
Z	6	14	23	26	27	28	29	30
Abs. edge	«1 keV	1.8 keV	5.5 keV	7.1 keV	7.7 keV	8.3 keV	9.0 keV	9.7 keV
$\Delta f'$	0.017	0.244	0.035	-1.179	-2.464	-2.956	-2.0255	-1.6142
$\Delta f''$	0.009	0.330	2.110	3.204	3.608	0.509	0.5885	0.6774

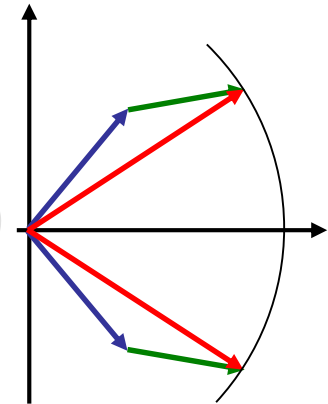
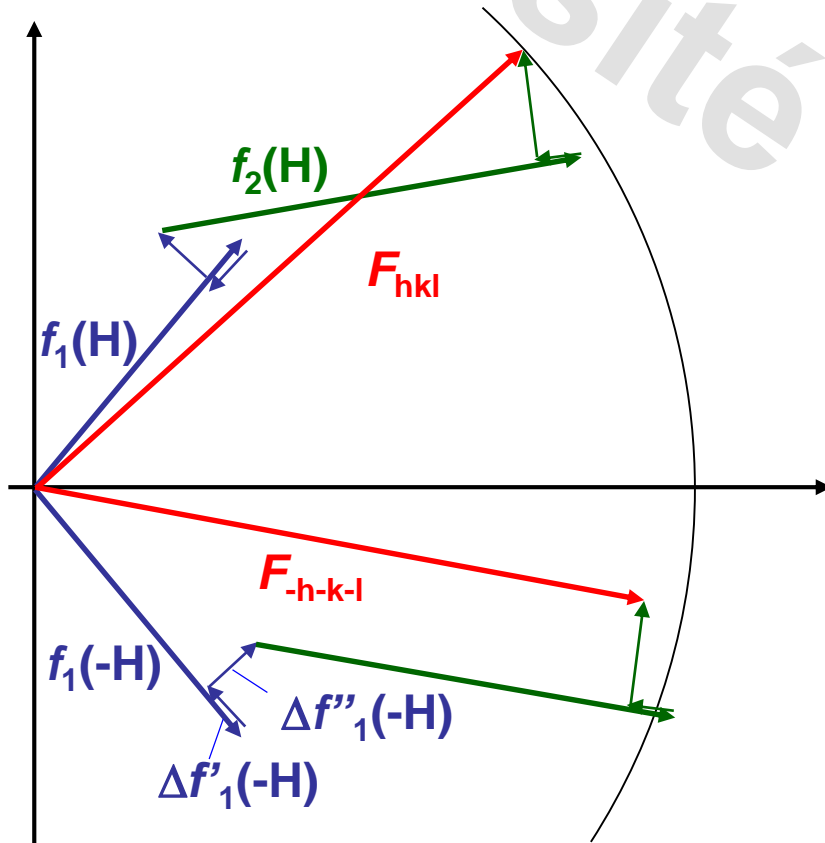
Cu K $\alpha$  radiation = 8.014 keV

from the next  
absorption edge



# Anomalous dispersion – non-centrosymmetric

- We can describe this effect with two correction factors:  $f = f_{\text{atomique}} + \Delta f' + i \cdot \Delta f''$ .
- Close to an absorption edge,  $\Delta f'$  becomes negative and can be related to X-ray absorption. The imaginary  $\Delta f''$  is positive at a phase 90° from that of  $f$ .
- As a consequence, **the Friedel law is not valid anymore!**



$$|F_{hkl}|^2 \neq |F_{\bar{h}\bar{k}\bar{l}}|^2$$

# Anomalous dispersion – non-centrosymmetric

$$F_{hkl} = \sum_{j=1}^N (f_j + \Delta f'_j + i \cdot f''_j) \cdot e^{2\pi i(h \cdot x_j + k \cdot y_j + l \cdot z_j)} = \sum_{j=1}^N (f'_j + i \cdot f''_j) \cdot e^{i\varphi_j}$$

$$= \sum_{j=1}^N f'_j (\cos \varphi_j + i \cdot \sin \varphi_j) + i \cdot f''_j (\cos \varphi_j + i \cdot \sin \varphi_j)$$

$$= \sum_{j=1}^N f'_j \cos \varphi_j - f''_j \sin \varphi_j + i \cdot \sum_{j=1}^N f''_j \cos \varphi_j + f'_j \sin \varphi_j$$

$$F_{\bar{h}\bar{k}\bar{l}} = \sum_{j=1}^N f'_j \cos(-\varphi_j) - f''_j \sin(-\varphi_j) + i \cdot \sum_{j=1}^N f'_j \sin(-\varphi_j) + f''_j \cos(-\varphi_j)$$

$$= \sum_{j=1}^N f'_j \cos \varphi_j + f''_j \sin \varphi_j + i \cdot \sum_{j=1}^N f''_j \cos \varphi_j - f'_j \sin \varphi_j$$

$$\left| F_{hkl} \right|^2 \neq \left| F_{\bar{h}\bar{k}\bar{l}} \right|^2$$

# Anomalous dispersion – centrosymmetric

$$F_{hkl} = \sum_{j=1}^N (f'_j + i \cdot f''_j) \cdot e^{i\varphi_j}; \quad \varphi_j = hx_j + ky_j + lz_j$$

**Inversion center :** For each atom  $j$  with  $x,y,z$  and phase  $\varphi$ , there is an atom with  $-x,-y,-z$  and thus phase  $-\varphi$ .

$$\begin{aligned} F_{hkl} &= \sum_{j=1}^{N/2} (f'_j + i \cdot f''_j) \cdot e^{i\varphi_j} + (f'_j + i \cdot f''_j) \cdot e^{-i\varphi_j} \\ &= \sum_{j=1}^{N/2} (f'_j \cos \varphi_j - f''_j \sin \varphi_j) + i \cdot (f'_j \sin \varphi_j + f''_j \cos \varphi_j) \\ &\quad + (f'_j \cos -\varphi_j - f''_j \sin -\varphi_j) + i \cdot (f'_j \sin -\varphi_j + f''_j \cos -\varphi_j) \\ &= \sum_{j=1}^{N/2} (f'_j \cos \varphi_j - f''_j \sin \varphi_j) + i \cdot (f'_j \sin \varphi_j + f''_j \cos \varphi_j) \\ &\quad + (f'_j \cos \varphi_j + f''_j \sin \varphi_j) + i \cdot (-f'_j \sin \varphi_j + f''_j \cos \varphi_j) \\ &= \sum_{j=1}^{N/2} 2f'_j \cos \varphi_j + i \cdot 2f''_j \cos \varphi_j \end{aligned}$$

$$\varphi_{j,\bar{h}\bar{k}\bar{l}} = (-hx_j - ky_j - lz_j) = -\varphi_{j,hkl} \Rightarrow$$

$$|F_{hkl}|^2 = |F_{\bar{h}\bar{k}\bar{l}}|^2$$

# Anomalous dispersion– wavelength dependance

Anomalous dispersion can normally be observed experimentally when the wavelength is close to an absorption edge (lisière d'absorption), i. e. the energy of the X-ray photon is close to an electronic excitation of one of the elements present.

	Cu K $\alpha$ (1.54 Å)			Mo K $\alpha$ (0.72 Å)			Ga K $\alpha$ (1.34 Å)		
	$\Delta f'$	$\Delta f''$	$\mu/\rho$	$\Delta f'$	$\Delta f''$	$\mu/\rho$	$\Delta f'$	$\Delta f''$	$\mu/\rho$
C	0.018	0.009	5	0.003	0.002	0.56	0.013	0.007	2
O	0.049	0.032	12	0.011	0.006	1.2	0.036	0.024	5
Co	-2.365	3.614	321	0.349	0.972	41	-0.711	2.90	184
Ni	-3.003	0.509	49	0.339	1.112	47	-1.374	3.27	209
Cu	-1.965	0.589	52	0.320	1.265	49	-2.819	3.68	216

For Mo K $\alpha$  radiation the presence of one Ni atom is equivalent (with regard to anomalous dispersion and determination of the absolute configuration) to the presence of **500 carbon atoms**!

Anomalous dispersion for light atoms is stronger for Cu than for Mo radiation. Cu is the preferred choice for determining the absolute structure of organic compounds.

Note that the position of absorption edges differ for each radiation. While Co has a much higher absorption coefficient (and anomalous dispersion) for Cu than for Mo radiation, the values for Ni are nearly equivalent, since Ni is found just behind an absorption edge.

# Determination of absolute configurations

**Old method:** Compare the R-values of a structure with its inverted structure

**Today:** We use the Flack-x parameter, which refines to give the ratio of a crystal or its inversion twin which fits best the observed intensities.

$$|F(H, x)|^2 = (1 - x)|F(H)|^2 + x|F(-H)|^2$$

$x = 0$ : correct absolute configuration

$x = 1$ : inverted absolute configuration

$0 < x < 1$ : racemic twin

**Chiral space groups (P1, P2<sub>1</sub>2<sub>1</sub>2<sub>1</sub>, ...):**

- Either no symmetry elements or only rotations
- May contain optically pure compounds
- **Flack parameter has to be calculated**

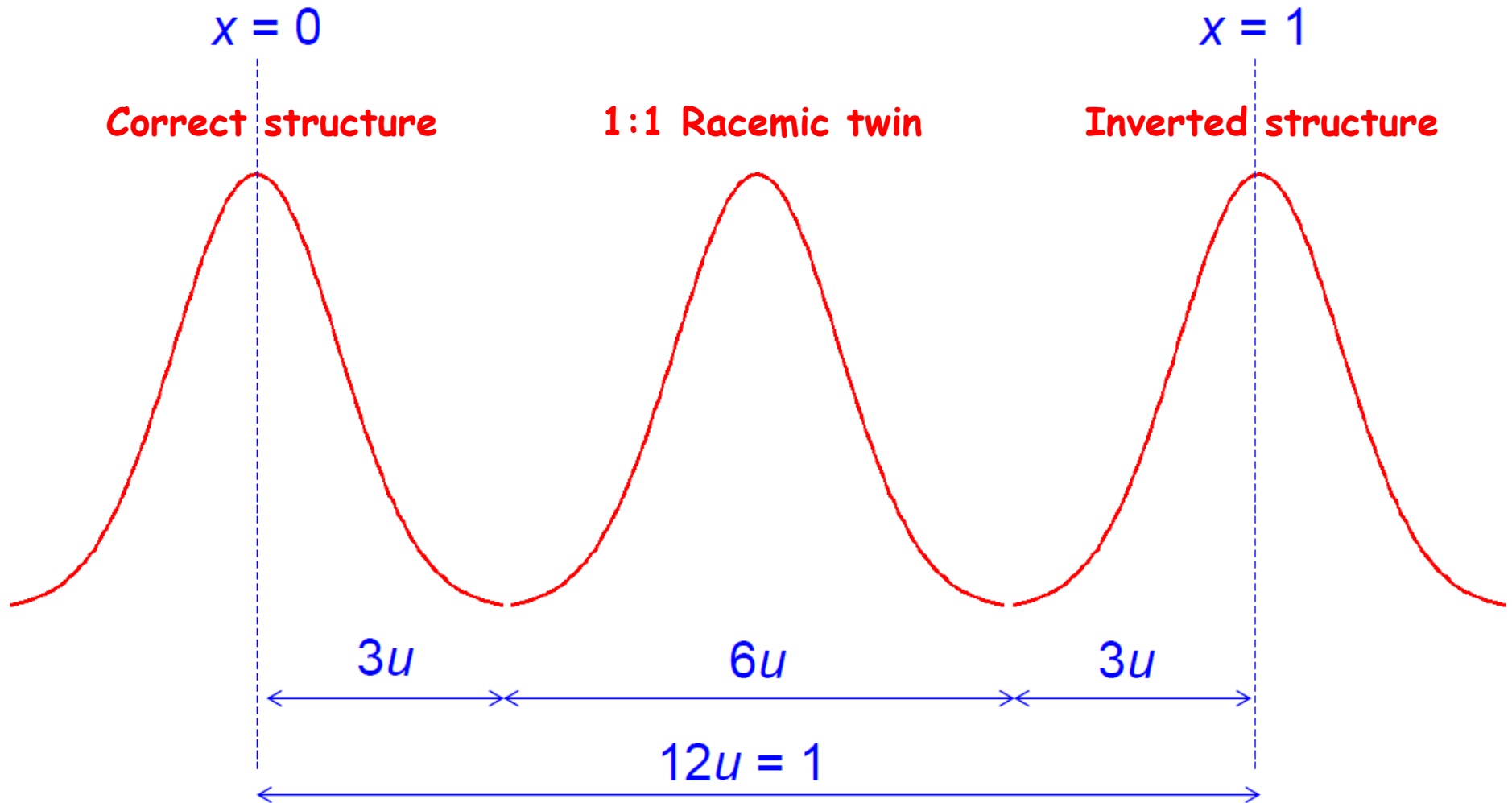
**Polar space groups (Pm, Pna2<sub>1</sub>, ...):**

- Contain mirror planes, but no inversion center
- May contain only achiral molecules or racemic mixtures
- **Flack parameter has to be calculated**

**Centrosymmetric space groups:**

- Flack parameter is not defined, since  $|F(H)|^2 = |F(-H)|^2$

# When is the Flack parameter reliable?



[http://www.gtecs.rwth-aachen.de/acmu/unterlagen/SParsons\\_Muelheim\\_2013.pdf](http://www.gtecs.rwth-aachen.de/acmu/unterlagen/SParsons_Muelheim_2013.pdf)

For the correct structure to be more likely than a racemic 1:1 twin, the Flack parameter should be  $0 \pm 3/12$ , i. e. the esd should be smaller than 0.1. **If the esd of the Flack parameter is  $>0.1$ , e. g. 0.02(14), the absolute structure cannot be determined.**

# Flack parameter calculation in SHELXL97

If the space group is non-centrosymmetric (chiral or polar), SHELXL97 estimates the Flack parameter by a fast refinement of the OSF (overall scale factor) and the Flack-parameter at the end of the refinement cycle. Only those two are refined against each other. This is undertaken to warn the user that he might have the wrong stereochemistry. **The obtained value is not the correct Flack parameter!**

To include the Flack parameter in the refinement, we have to introduce the following two commands in the ins-file:

```
TWIN  
BASF 0.4
```

TWIN without any other data assumes the presence of a racemic twin. The refinement of the **B**atch **S**cale **F**actor for a racemic twin is identical to the Flack parameter. Refinement without BASF – TWIN might yield wrong values, in particular for the standard deviation of the Flack parameter, such as  $x = 0.23(32)$ . **For non-centrosymmetric space groups TWIN – BASF 0.4 must be used during the refinement!**

# Flack parameter in SHELXL2014 / Parsons' quotient

Later work has shown that determination of the Flack parameters using quotients (**Parsons' method**) eliminates systematic errors in the dataset and yields lower uncertainties for the Flack parameter. (Parsons, S., Flack, H. D. & Wagner, T. *Acta Cryst.* **2013**, B69, 249)

The Flack  $x$  parameter is still defined according to

$$\begin{aligned} |F(H, x)|^2 &= (1-x)|F(H)|^2 + x|F(-H)|^2 \\ \Leftrightarrow I_{hkl}(x) &= (1-x)I_{hkl} + xI_{\bar{h}\bar{k}\bar{l}} \end{aligned}$$

Using now **quotients** of the **differences** of Friedel pairs, one obtains

$$\frac{I_{hkl}(x) - I_{\bar{h}\bar{k}\bar{l}}(x)}{I_{hkl}(x) + I_{\bar{h}\bar{k}\bar{l}}(x)} = (1-2x) \frac{I_{hkl} - I_{\bar{h}\bar{k}\bar{l}}}{I_{hkl} + I_{\bar{h}\bar{k}\bar{l}}}$$

**This can be measured:**  $I_{hkl}(x) = I_{obs}$

**This is your model:**  $I_{hkl} = I_{calc}$

With  $Q = \frac{I_{hkl} - I_{\bar{h}\bar{k}\bar{l}}}{I_{hkl} + I_{\bar{h}\bar{k}\bar{l}}}$  this can be written as  $Q_{obs} = (2-x)Q_{calc}$

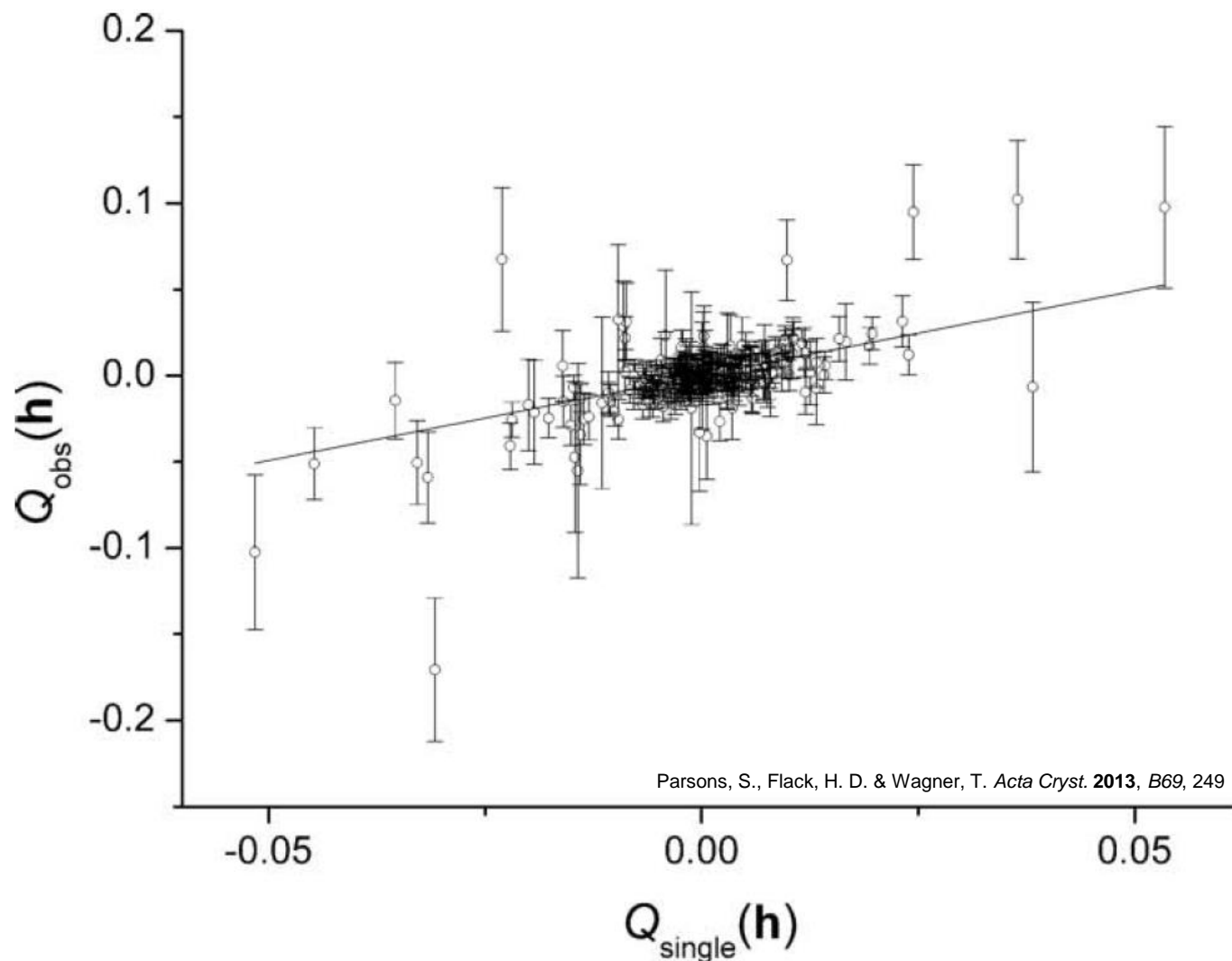


# The Flack parameter in SHELXL2014 / Parsons' quotient

According to  $Q_{obs} = (2 - x)Q_{calc}$

a plot of the quotient obtained from the measured intensities versus the quotient obtained from calculated intensities yields a line with a slope of  $2-x$ . Simple linear regression thus yields the Flack-parameter  $x$ .

(Sometimes also called Parsons'  $z$ .)



# The Flack parameter in SHELXL2014 / Parsons' quotient

Parsons, Flack and Wagner showed that the quotient method determines the Flack-parameter with **a lower statistical error** than the direct refinement.

Structure code	Chemical formula	Friedif <sub>stat</sub>	Space group	Redundancy	$R_1[ F  > 4\sigma( F )]$	$x$ (twin)	$y$ (Hoofit)	$x$ (quotient)	$x$ (difference)
<i>R</i> -Mandelic acid <sup>A</sup>	C <sub>8</sub> H <sub>8</sub> O <sub>3</sub>	36	$P2_1$	11	0.0511	0.12 (46)	0.03 (6)	0.00 (11)	−0.04 (5)
L-Alanine <sup>B</sup>	C <sub>3</sub> H <sub>7</sub> NO <sub>2</sub>	34	$P2_12_12_1$	25	0.0219	−0.04 (27)	0.01 (4)	0.01 (4)	0.00 (9)
L-Alanine <sup>A</sup>	C <sub>3</sub> H <sub>7</sub> NO <sub>2</sub>	34	$P2_12_12_1$	15	0.0181	0.06 (26)	0.06 (5)	0.01 (3)	0.01 (3)
Glutamine <sup>B</sup>	C <sub>5</sub> H <sub>8</sub> N <sub>2</sub> O <sub>2</sub>	33	$P2_12_12_1$	28	0.0248	0.09 (25)	0.07 (3)	0.05 (4)	0.04 (3)
GKO02 <sup>B</sup>	C <sub>25</sub> H <sub>31</sub> NO <sub>5</sub>	32	$P2_12_12_1$	15	0.0247	0.01 (15)	0.03 (3)	0.08 (4)	0.07 (4)
A0030a <sup>B</sup>	C <sub>21</sub> H <sub>26</sub> N <sub>2</sub> O <sub>3</sub>	29	$P2_12_12_1$	11	0.0263	−0.10 (21)	−0.07 (5)	0.07 (3)	0.04 (2)
A0034a <sup>B</sup>	C <sub>18</sub> H <sub>25</sub> O <sub>2.5</sub>	29	$P2_12_12$	11	0.0274	0.00 (21)	0.02 (3)	0.09 (3)	0.07 (3)
A0034b <sup>B</sup>	C <sub>18</sub> H <sub>25</sub> O <sub>2.5</sub>	29	$P2_12_12$	35	0.0268	−0.01 (22)	−0.02 (3)	0.02 (3)	0.02 (2)
LRE01a <sup>B</sup>	C <sub>14</sub> H <sub>19</sub> NO	24	$P2_12_12_1$	22	0.0278	−0.01 (33)	−0.03 (5)	0.02 (2)	0.06 (2)
TWA18a <sup>B</sup>	C <sub>16</sub> H <sub>20</sub> N <sub>2</sub> O	23	$P2_12_12_1$	17	0.0253	0.04 (26)	0.04 (3)	−0.02 (2)	−0.01 (3)
								−0.02 (2)	−0.02 (2)
								−0.04 (6)	−0.04 (3)
								−0.02 (5)	−0.03 (5)
								0.07 (3)	0.00 (2)
								0.04 (3)	0.04 (3)

Parsons, S., Flack, H. D. & Wagner, T. *Acta Cryst.* **2013**, B69, 249

They also showed that this determination is stable as a **post-refinement method**, i. e. the Flack parameter does **not** have to be included in the refinement cycle. Thus, determination of the correct Flack parameter using Parsons' quotient does not require a BASF/TWIN command anymore. The latter should still be used, however, if a racemic twin is indeed present.

# The Hooft parameter $y$

A different approach to determine the Flack parameter uses Bayesian statistics, i. e. the determination of the probability that you have the correct structure.

- R. W. W. Hooft, L. H. Straver, A. L. Spek *J. Appl. Cryst.* **2008**, 41, 96 and *J. Appl. Cryst.* **2010**, 43, 665

**Friedel pair:** a pair of reflexions related by inversion symmetry, i. e.  $I_{hkl}$  and  $I_{(-h)(-k)(-l)}$ .

**Bijvoet pair:** a pair of reflexions, whose true symmetry equivalents are Friedel pairs.

E. g. for  $Pm$  we have

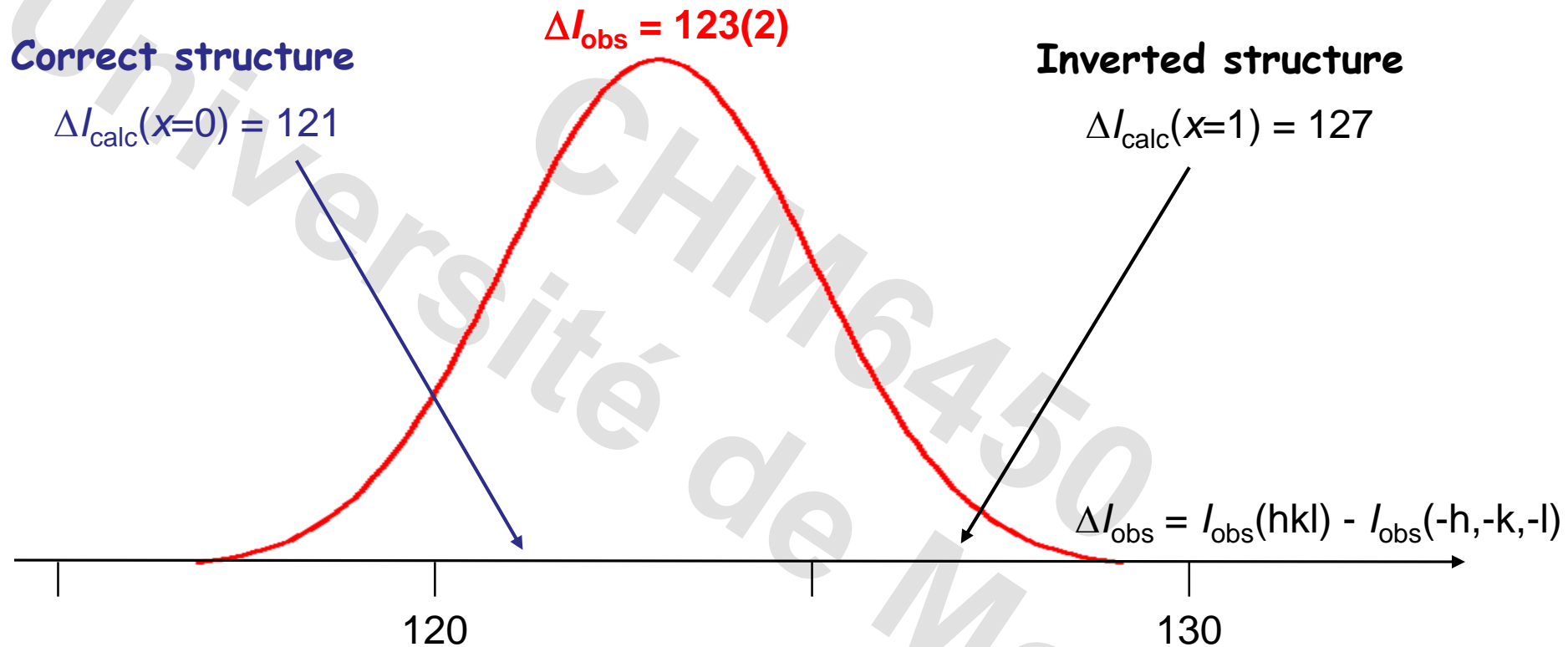
$$I_{hkl} = I_{h-k-l} \quad \text{and} \quad I_{-h-k-l} = I_{-hk-l}$$

We thus have the following Bijvoet pairs:

$$I_{hkl} \mid I_{-h-k-l}, \quad I_{hkl} \mid I_{-hk-l}, \quad I_{h-k-l} \mid I_{-h-k-l}, \quad I_{h-k-l} \mid I_{-hk-l}.$$

The Friedel law (and respective anomalous dispersion differences) apply to Bijvoet pairs in the same way as to Friedel pairs. Since Friedel pairs are always Bijvoet pairs, but not all Bijvoet pairs are Friedel pairs, we have (above the triclinic space groups) always more Bijvoet pairs than Friedel pairs.

# The Hooft parameter $y$



Observed reflection:  $\Delta I_{\text{hkl}} = 123(2)$ . For the model, we calculate  $\Delta I_{\text{calc}} = 121$ , for the inverted model  $\Delta I_{\text{calc}} = 127$ . The probability that the model is correct **for this reflection** can be expressed by

$$p_0 = \frac{1}{\sqrt{2\pi}} e^{-\frac{\left(\frac{\Delta I_{\text{calc}} - \Delta I_{\text{obs}}}{\text{esd}(\Delta I_{\text{obs}})}\right)^2}{2}} = \frac{1}{\sqrt{2\pi}} e^{-\frac{(2/2)^2}{2}} = 24\%$$

The respective calculation for the inverted model yields  $p_1 = 5\%$

# The Hooft parameter $y$

The probability that the whole model is correct, is obtained by multiplication of all probabilities of the observed Bijvoet differences:

$$p(x=0) = \prod p_0 \quad \text{with} \quad p_0 = \frac{1}{\sqrt{2\pi}} e^{-\frac{\left(\frac{\Delta I_{calc}(x=0) - \Delta I_{obs}}{esd(\Delta I_{obs})}\right)^2}{2}}$$

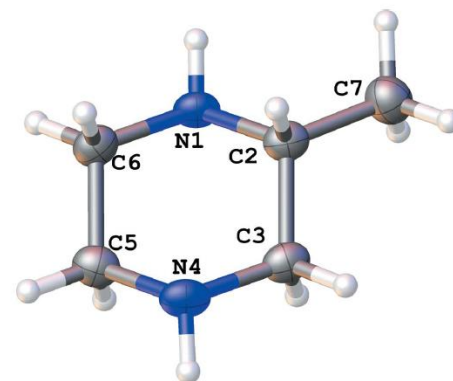
$$p(x=1) = \prod p_1 \quad \text{with} \quad p_1 = \frac{1}{\sqrt{2\pi}} e^{-\frac{\left(\frac{\Delta I_{calc}(x=1) - \Delta I_{obs}}{esd(\Delta I_{obs})}\right)^2}{2}}$$

Some statistics follows (look up the 2008 paper): Probability  $p$  is calculated for a continuum of  $x = 0..1$  with Bayesian statistics and the result interpreted as a Gaussian probability distribution, centered around the **Hooft parameter  $y$**  (analogous to  $x$ ).

In case of poor data with systematic errors, the error distribution is not Gaussian. In 2010 Hooft *et al.* improved their method by using a student-t distribution instead of a Gaussian distribution to interpret their obtained probability distribution.

# Comparison of the methods

J. Reibenspiess, N. Bhuvanesh  
*Acta Cryst. B*, **2013** B69, 288



*R*-2-methylpiperazine

Comparison of absolute structure parameters.

	Flack (TWIN) <sup>†</sup>	Flack (Post) <sup>‡</sup>	Parsons' Q <sup>§</sup>	Hooft-G <sup>¶</sup>	Hooft-S <sup>††</sup>
Rmpip	0.216 (871)	0.214 (832)	0.09 (23)	0.3 (3)	0.09 (8)
Smpip	−0.163 (826)	−0.160 (804)	0.07 (24)	0.2 (3)	0.05 (8)
RmpipBr	0.03 (4)	0.03 (4)	0.04 (1)	0.04 (1)	0.05 (1)
SmpipBr	0.06 (4)	0.06 (4)	0.04 (1)	0.04 (2)	0.03 (1)

Rmpip	0.216 (871)	0.214 (832)	0.09 (23)	0.3 (3)	0.09 (8)
Smpip	−0.163 (826)	−0.160 (804)	0.07 (24)	0.2 (3)	0.05 (8)
RmpipBr	0.03 (4)	0.03 (4)	0.04 (1)	0.04 (1)	0.05 (1)
SmpipBr	0.06 (4)	0.06 (4)	0.04 (1)	0.04 (2)	0.03 (1)

<sup>†</sup> *SHELXL12* (Sheldrick, 2012) TWIN-1000-1000-1/BASF parameter refined (Flack, 1983). <sup>‡</sup> *SHELXL12* (Sheldrick, 2012) post-refinement Flack parameter determination (Flack, 1983). <sup>§</sup> *SHELXL12* (Sheldrick, 2012) Parsons' Q parameter determination (Parsons & Flack, 2004). <sup>¶</sup> *PLATON* (Spek, 2012), Version 161012, Hooft parameter determination, Gaussian distribution (Hooft *et al.*, 2008). <sup>††</sup> *PLATON* (Spek, 2012), Version 161012, Hooft parameter determination, student-T distribution (Hooft *et al.*, 2009).

# How to calculate these values and how to invert a structure

- **SHELX2012** will automatically calculate the Flack parameter using Parsons' quotient.
- **OLEX2** will calculate the Flack parameter using Parsons' quotient and the Hooft y parameter. (For student distribution, go to INFO - Bijvoet Differences Probability Plot)
- **Platon** will calculate the Flack parameter using Parsons' quotient (also called Parsons' z) and Hooft for Gaussian and student distribution.

## How to invert the structure if the Flack parameter refines to 1:

- Either use the command "inv -f" in OLEX (**my preferred choice**)
- or introduce the command MOVE 1 1 1 -1 in the ins file

**Attention:** For enantiomorphic space groups, inversion of the structure requires changement of the space group.

**Attention:** There are a few cases, where inverting the structure at the origin does **not** work. These are (see SHELX manual):

Fdd2	MOVE .25 .25 1 -1	I4 <sub>1</sub> cd	MOVE 1 .5 1 -1
I4 <sub>1</sub>	MOVE 1 .5 1 -1	I-42d	MOVE 1 .5 .25 -1
I4 <sub>1</sub> 22	MOVE 1 .5 .25 -1	F4 <sub>1</sub> 32	MOVE .25 .25 .25 -1
I4 <sub>1</sub> md	MOVE 1 .5 1 -1		

## **If the absolute structure is important**

- Higher intensities normally equate lower errors in the Flack parameter. Thus try to choose the biggest crystal.
- Redetermination with a second crystal lowers the chance that you picked the wrong enantiomer by chance.
- Single crystals chosen from a high-yield re-crystallization are more reliable than to use the two single crystals which were the only ones crystallizing.
- In particular if crystallization yield is low, measure the powder diagram of your bulk sample and compare to the one calculated from your structure determination to exclude the presence of two phases.



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# **Special cases**

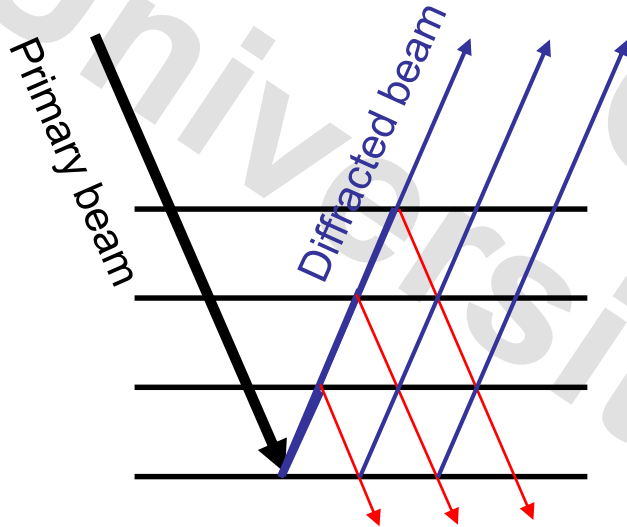
**Anomalous dispersion**

**Extinction**

**Violation of systematic absences**

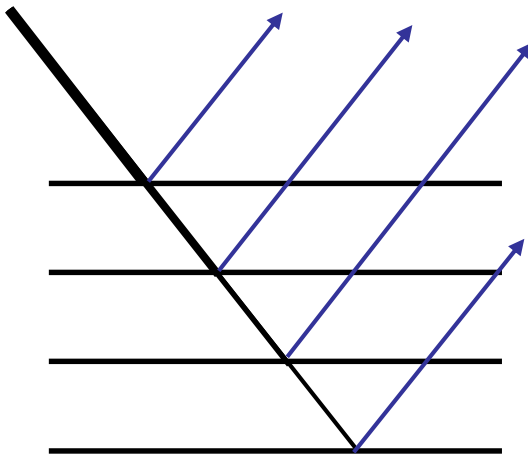
**If things went wrong...**

# Extinction



## Primary extinction

- **Weakening of the diffracted beam** due to repeated diffraction (aggravated by destructive interaction with double-diffracted beams)
- Only in perfect crystals
- Difficult to describe (dynamic scattering)
- Might reduce  $I \sim |F|^2$  down to  $I \sim |F|$  in extreme cases



## Secondary extinction

- Weakening of the primary beam due to precedent diffractions
- Particularly important in perfect crystals
- Comparable to absorption effects

# How to correct extinction during the refinement

**Warning signs of extinction:**  $|F_{\text{obs}}| < |F_{\text{cal}}|$  for reflections with high intensity. SHELXL (XL) will print a warning, should refinement of an extinction correction be required (.lst file and screen output during refinement).

**Extinction treatment in SHELXL:** The command **EXTI [x]** refines an extinction parameter x. Extinction correction is done by the following formula:

$$F_c^{\text{corr.}} = F_c \left( 1 + \frac{x F_c^2 \lambda^3}{\sin 2\theta} \right)^{-1/4}$$

This correction is purely empiric, but it works reasonably well.

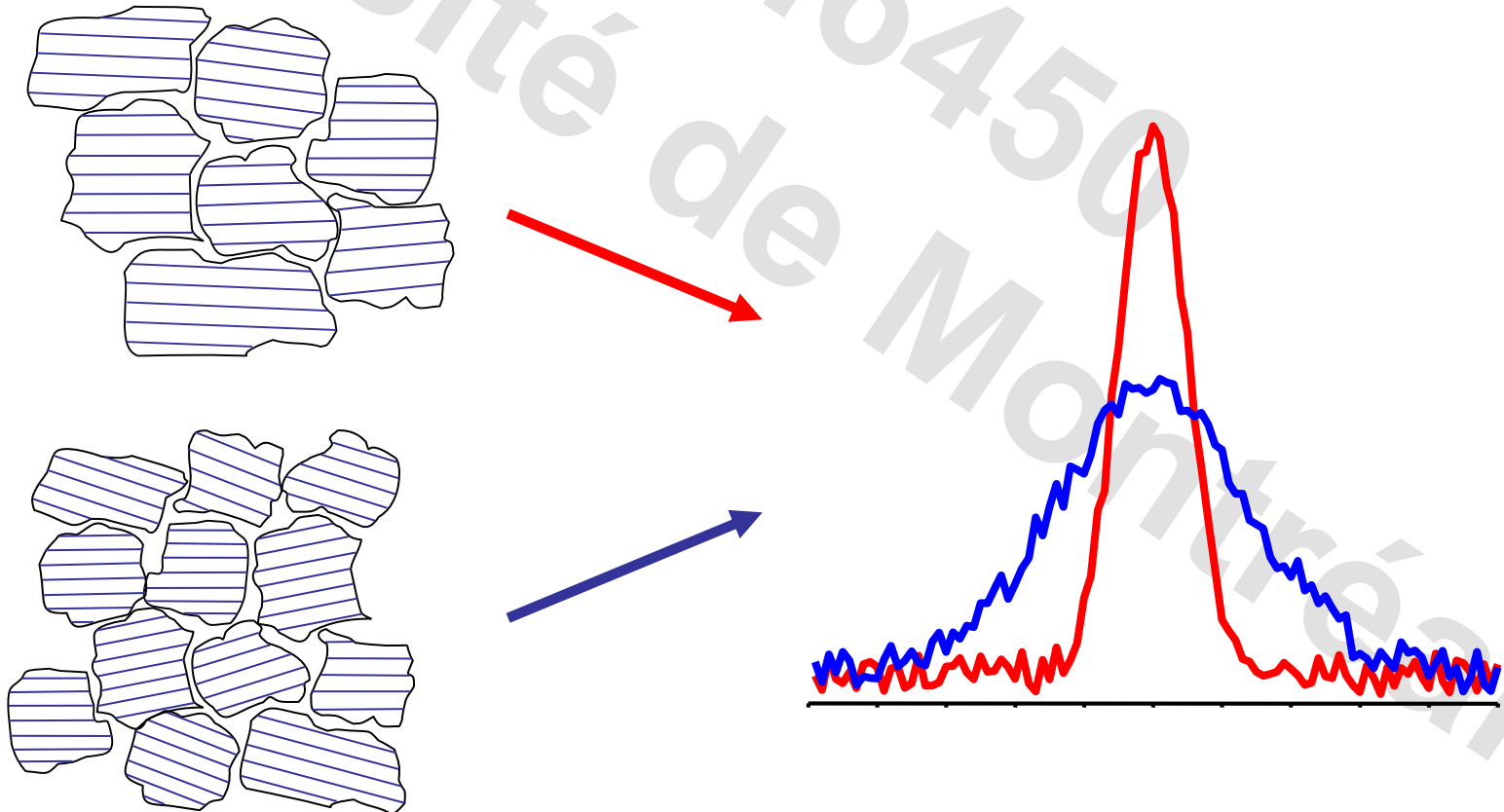
**Attention:** There are a lot of other effects which will cause  $|F_{\text{obs}}| < |F_{\text{cal}}|$ ! The EXTI command should thus be introduced only as the last step of the refinement (after hydrogen atoms etc.), when still needed. Should the extinction factor refine to 0 (in the margin of its error), you should better remove it from the refinement.

We can reduce the extinction by increasing the mosaic structure of our crystals, i. e. by dipping them shortly in liquid nitrogen. **This is not a standard procedure for molecular crystals!** (In other words: do not do this to your crystals.)

# Mosaic structure

Perfect long-range crystalline order is observed only in special cases, for example in purified superconductor materials. “Real” crystals have a mosaic structure: only small domains show perfect crystalline order, but these perfect domains are slight offset from each other by  $1\text{-}2^\circ$ .

The mosaic structure is the main reason for peak broadening in X-ray diffraction.



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# **Special cases**

**Anomalous dispersion**

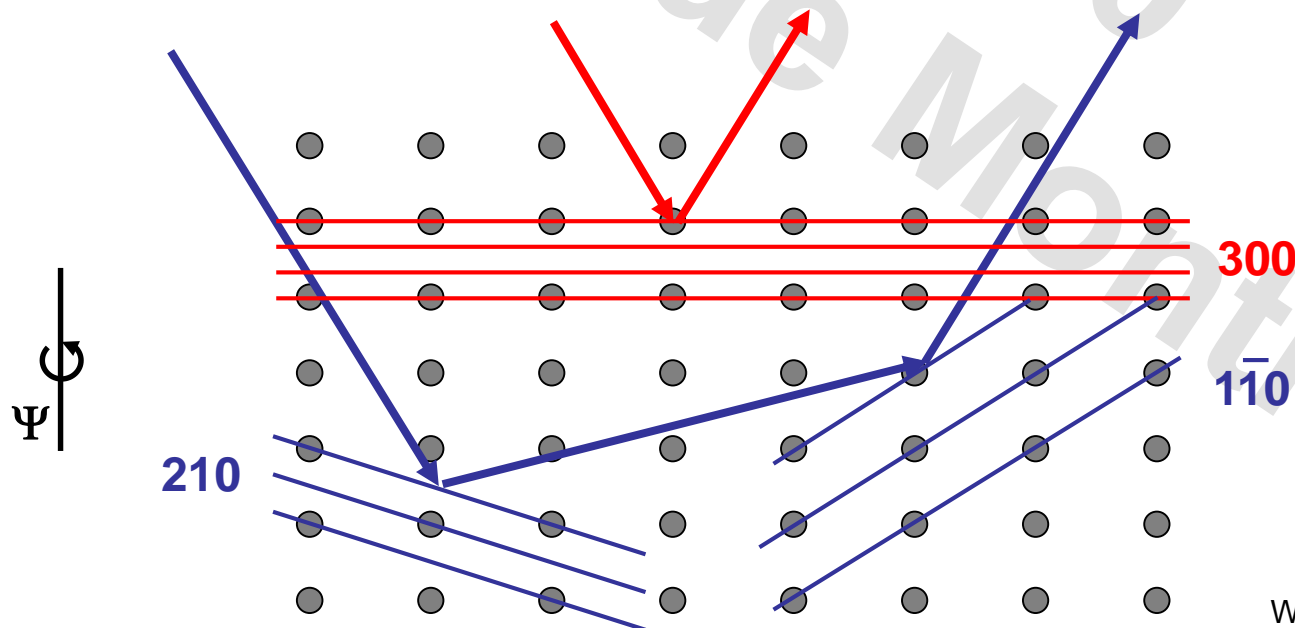
**Extinction**

**Violation of systematic absences**

**If things went wrong...**

# Renninger effect

- One reason why systematic absences might be violated
- A diffraction at plane  $hkl$  is obtained by double diffraction at  $h'k'l'$  and  $(h-h')(k-k')(l-l')$ .
- The Renninger effect is noticeable only, when the two reflections  $h'k'l'$  and  $(h-h')(k-k')(l-l')$  are strong and the reflection  $hkl$  is weak (e. g. systematically absent)
- Rotation around  $\Psi$  (**perpendicular to the  $hkl$  plane**) moves planes  $h'k'l'$  and  $(h-h')(k-k')(l-l')$  out of their reflection condition and destroys the Renninger effect.



# $\lambda/2$ effect

The majority of monochromators are based on interference, i. e. diffraction, to monochromize X-ray light. Due to the Bragg law

$$2d_{hkl} \sin \theta = n\lambda$$

we find reflections of the order  $n$  of light with wavelengths  $\lambda/n$  at the same  $\theta$  angle.

$K\alpha$  radiation of the wavelength  $\lambda$  is thus always accompanied by a small percentage ( $< 0.3\%$ ) of radiation with the wavelength  $\lambda/2$ .

A reflection  $hkl$  is thus always superimposed with a reflection  $2h,2k,2l$  for  $\lambda/2$ .

This is normally not a problem due to the small intensity of the  $\lambda/2$  radiation ( $< 0.3\% \lambda/2$ ), but it becomes noticeable **when the reflection  $2h,2k,2l$  is strong and the reflection  $hkl$  weak, i. e. systematically absent.**

**Correction:** We normally do not correct for  $\lambda/2$  effects. If we want to, we have to determine the amount of  $\lambda/2$  radiation using a (perfect) standard crystal.

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# **Special cases**

**Anomalous dispersion**

**Extinction**

**Violation of systematic absences**

**If things went wrong...**



# My R-value is too high...

## What is too high?

Compare  $R_1$  to  $R_\sigma$ . (in the CIF  $R_\sigma = \text{\_diffn\_reflns\_av\_unetl/netl}$ )

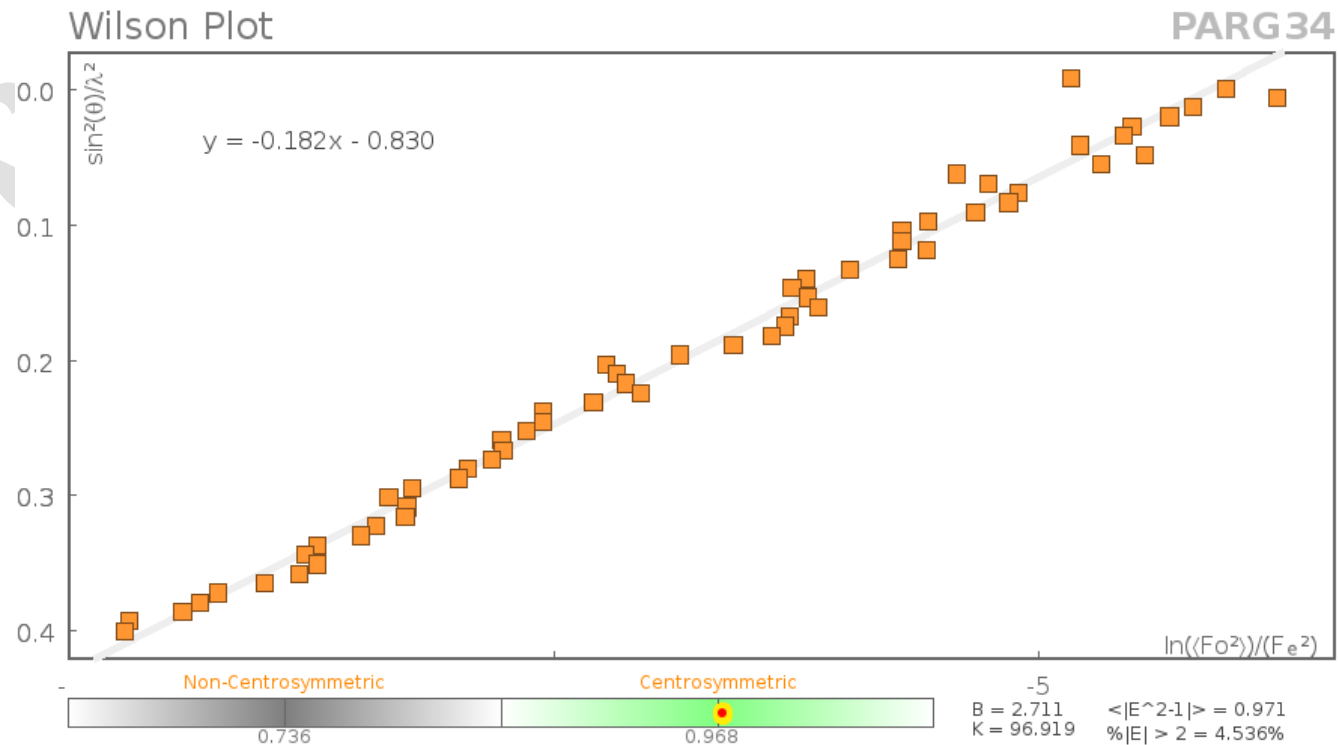
- If  $R_1 < R_\sigma$ , your problem is your crystal (signal/noise). Go back to the lab, re-crystallize and stop bothering crystallographers!
- If  $R_1 > R_\sigma$ , your problem is **not** the crystal. Try to find out where the problem is.

# Verification tools in OLEX

Olex2 offers under INFO – Reflection Statistics several plots, which allow us to evaluate our reflection data. While worthwhile looking at, we normally do not have to bother if R1 is well below 5% or if R1 is well below  $R\sigma$ .

## Wilson plot

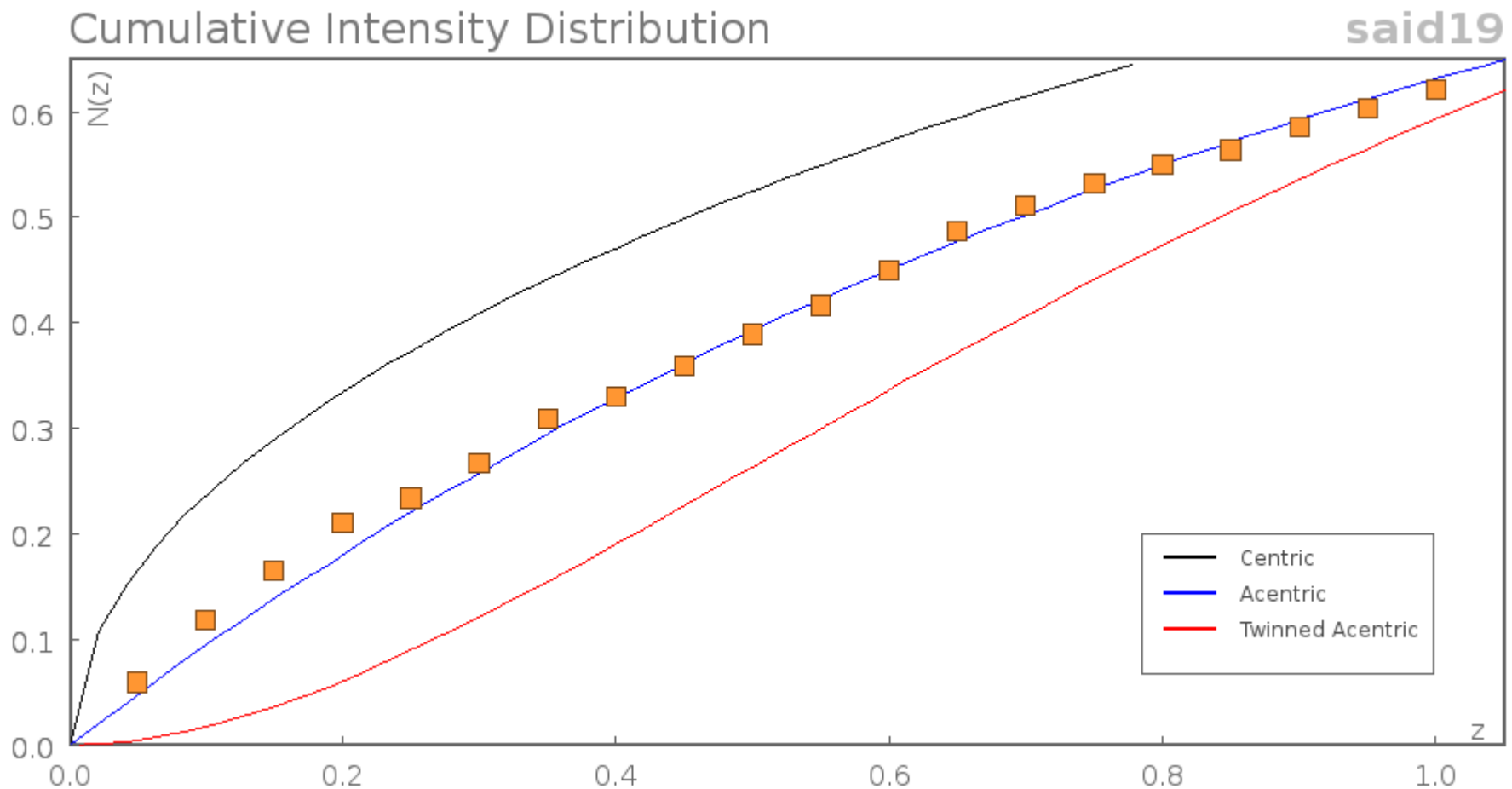
The Wilson plot shows the observed intensities plotted against those expected for a random distribution of atoms. It should be (roughly) a straight line for small molecular crystallography.



Strong variation from a straight line indicate areas of high order and less order (large amounts of disordered solvent). From the slope of the line we can determine the overall temperature factor B, which should be  $<3$  for a low temperature structure in the absence of strong disorder.

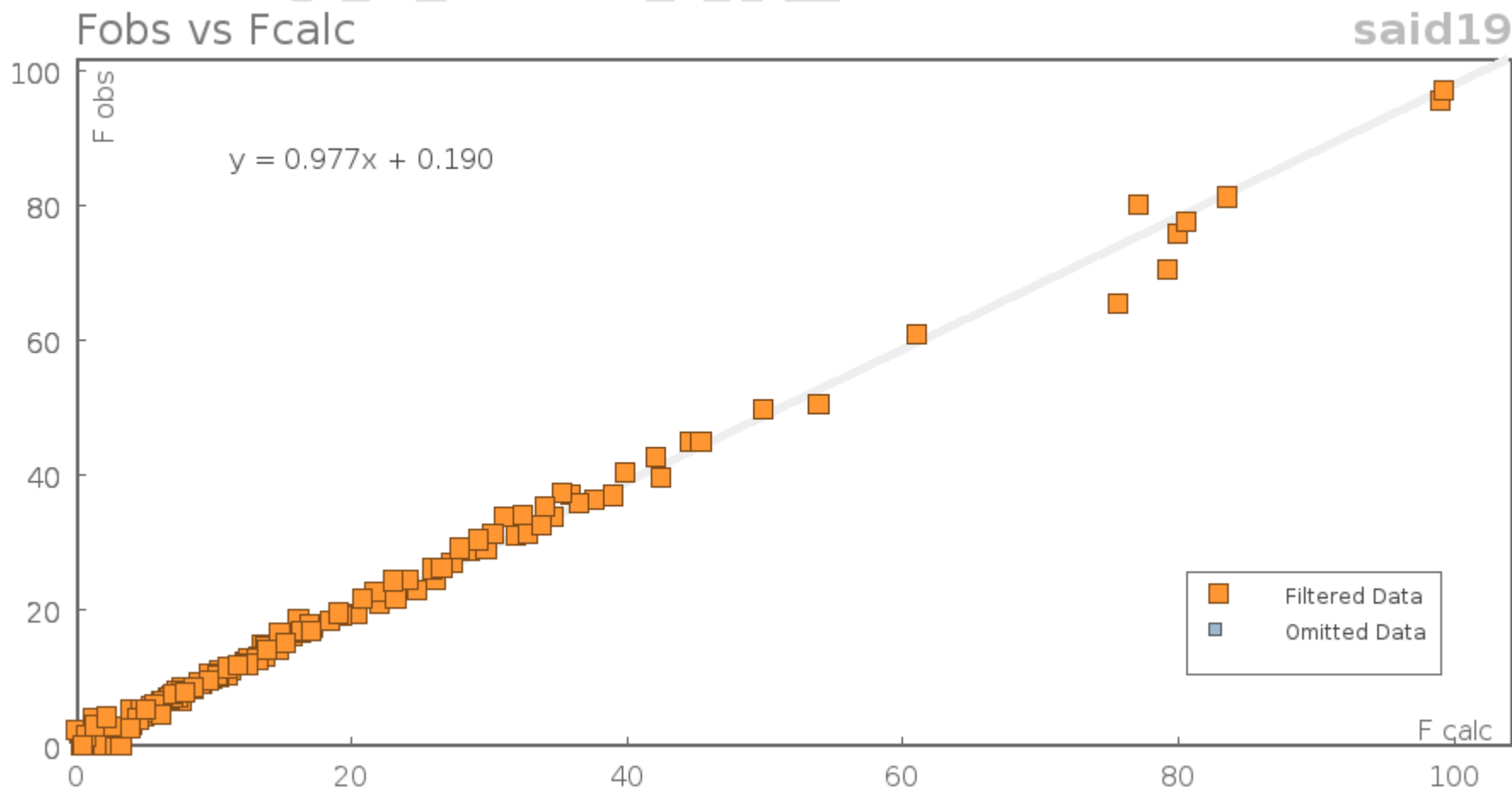
# Verification tools in OLEX - Cumulative Intensity

This shows the distribution of (normalized) intensities (E-values). Typically centered structures have a larger probability to have very intense or very weak reflections. Comparison of the observed intensities with theoretical curves indicates if a structure is centric or acentric. Deviations from the expected distribution might indicate problems.



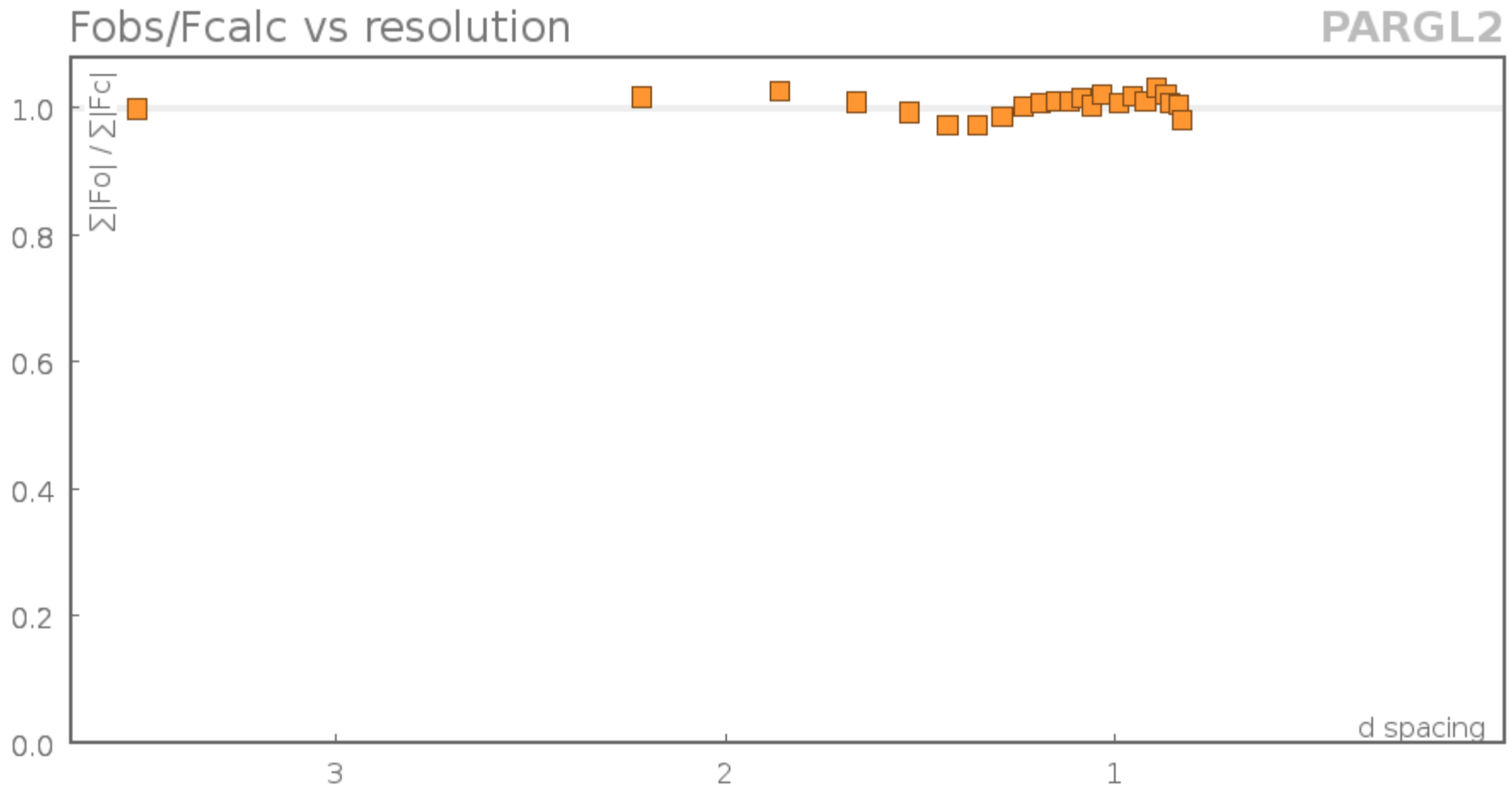
# Verification tools in OLEX – $F_{\text{obs}}$ vs. $F_{\text{calc}}$

Plotting observed vs. calculated structure factors should yield a straight line with a slope of 1. Outliers are easily identified (hover over the data point with the mouse) and indicate problems with these reflections (-> OMIT). A curvature or slopes larger or smaller than 1 might indicate systematic problems (absorption, cutting of too intense reflections, extreme disorder, twinning...)



# Verification tools in OLEX – $F_{\text{obs}}/F_{\text{calc}}$

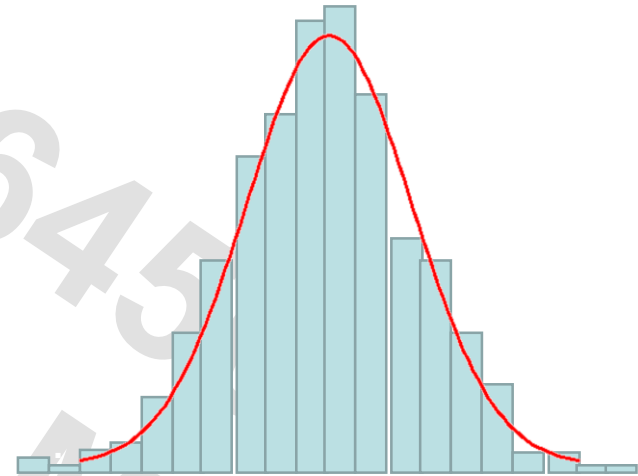
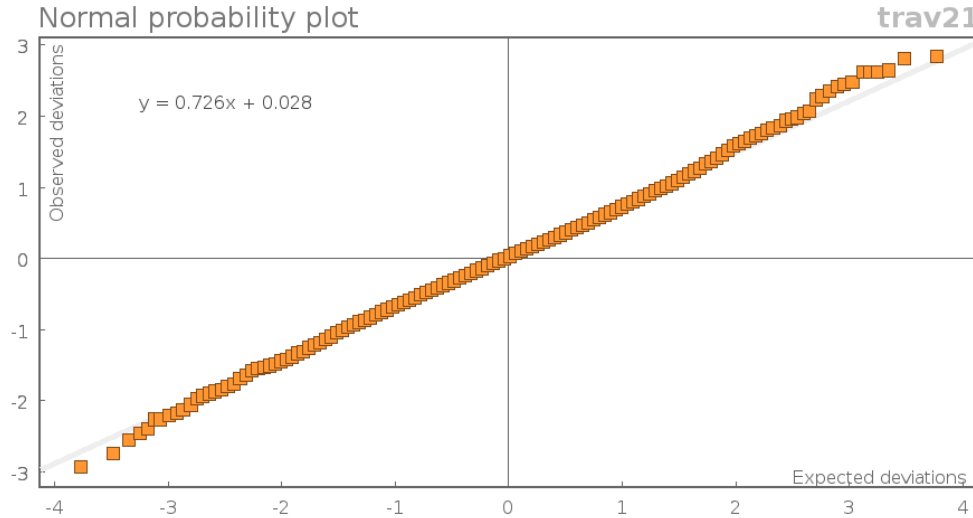
Similar to  $F_{\text{obs}}$  vs.  $F_{\text{calc}}$ , but deviations are easier to see. The ratio  $F_{\text{obs}}/F_{\text{calc}}$  can be plotted against  $d$ -spacing,  $\theta$  or  $\sin \theta / \lambda$ . It should be 1 with minor variations. A systematic variation of  $F_{\text{obs}}/F_{\text{calc}}$  in dependence of the resolution indicates problems with the data set (insufficient absorption correction, excessive disorder, twinning, ...)



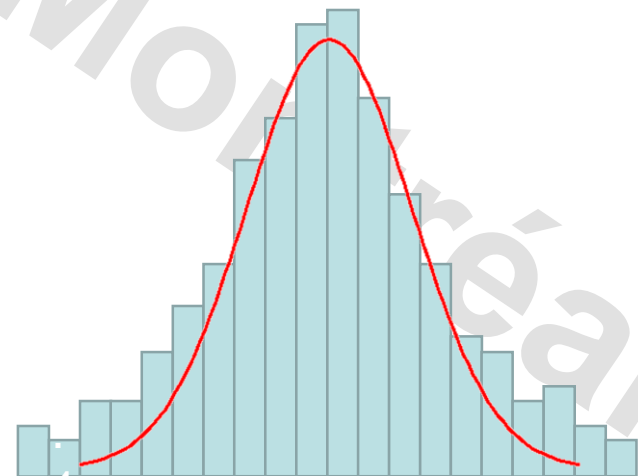
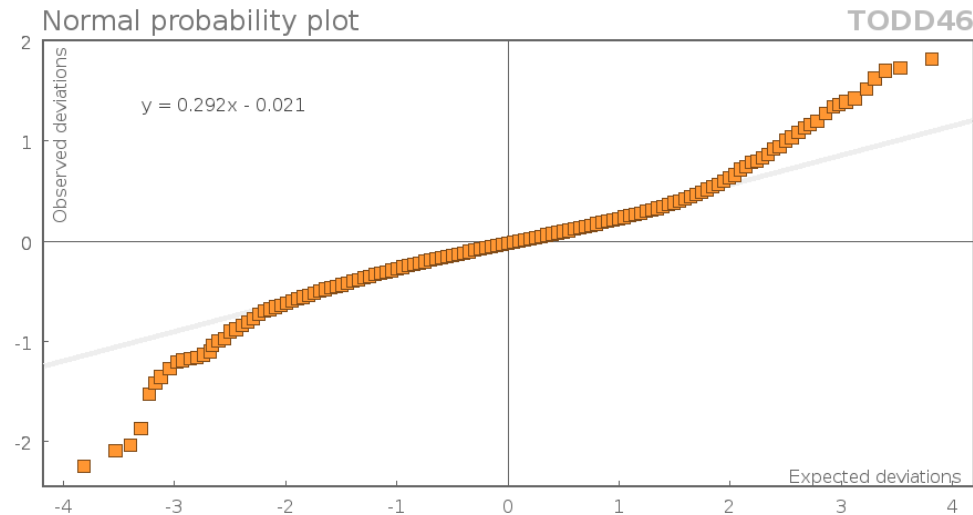
# Verification tools in OLEX – normal probability

The normal probability plot shows how well the observed differences  $F_{\text{obs}}/F_{\text{calc}}$  confirm to a Gaussian distribution expected for random errors. Systematic errors typically lead to deviations from linear behaviour. Even for good structures, this can often be seen. As long as the normal probability plot remains linear in the central part and symmetrical, this normally does not indicate a problem. Attention: the normal probability plot is highly sensitive towards the weighing scheme. Make sure to have refined it correctly.

Normal probability plot

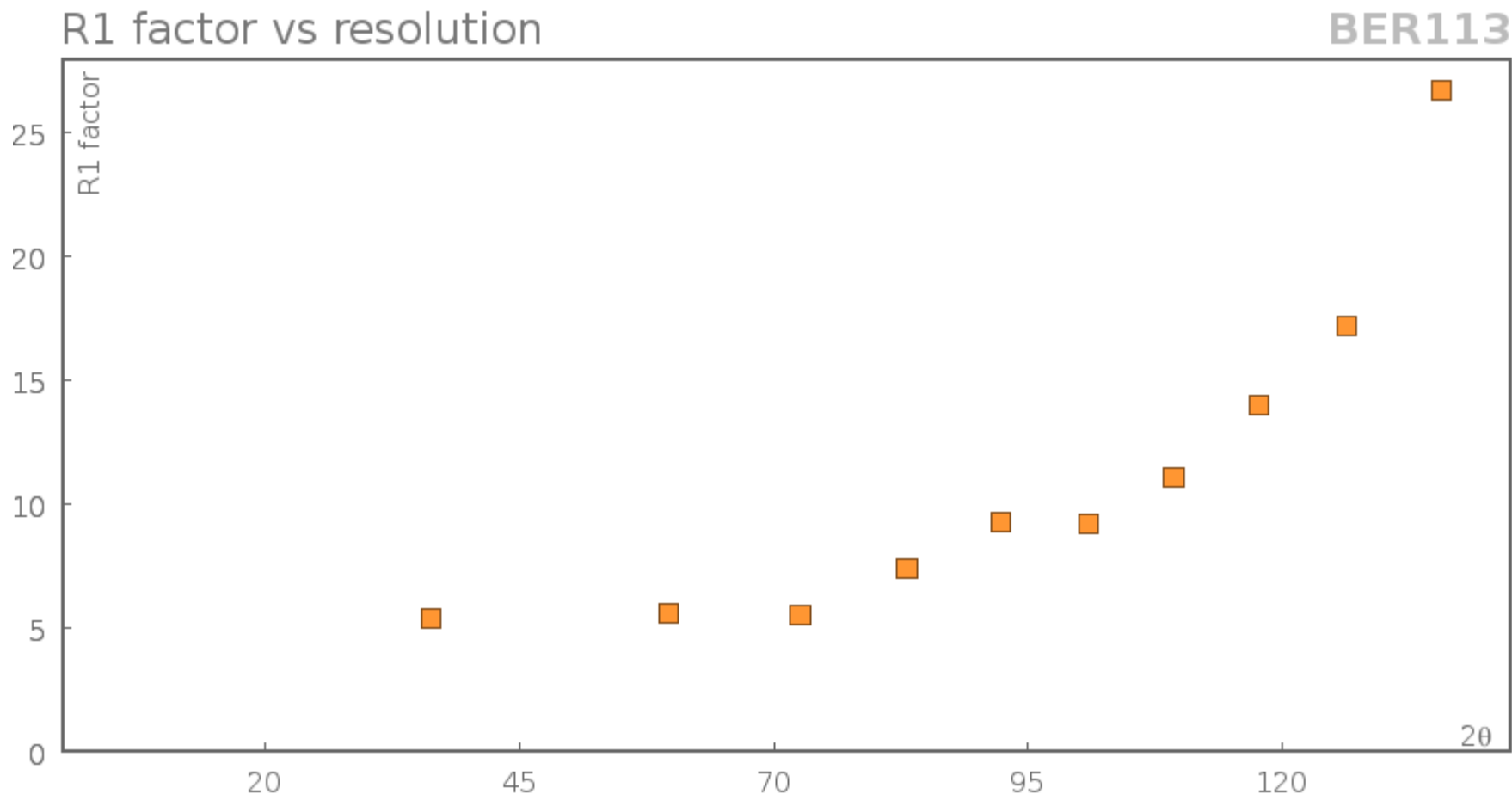


Normal probability plot



# Verification tools in OLEX – R1 vs. resolution

Due to the generally lower intensities of reflections at high  $\theta$  angle, the R-factor should increase steadily (in a slight exponential) with resolution. (OLEX can plot this either vs.  $\theta$ ,  $\sin \theta/\lambda$ , or d-spacing.) **Any maxima in this curve would indicate severe problems** with the data set which mostly likely effect a limited number of runs.

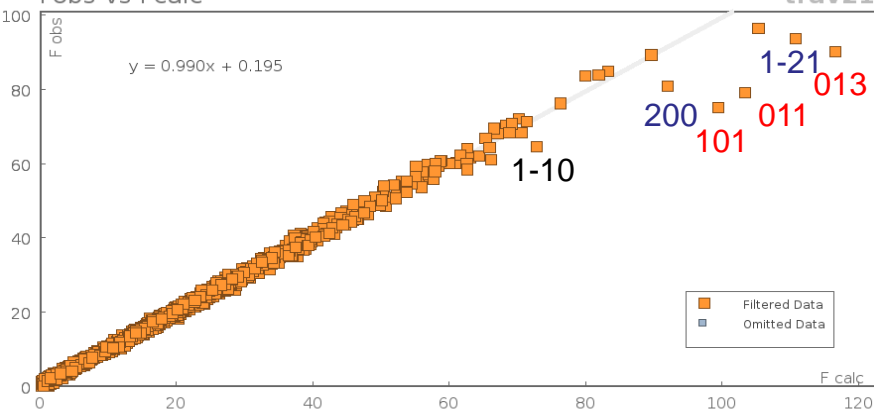


# Case examples

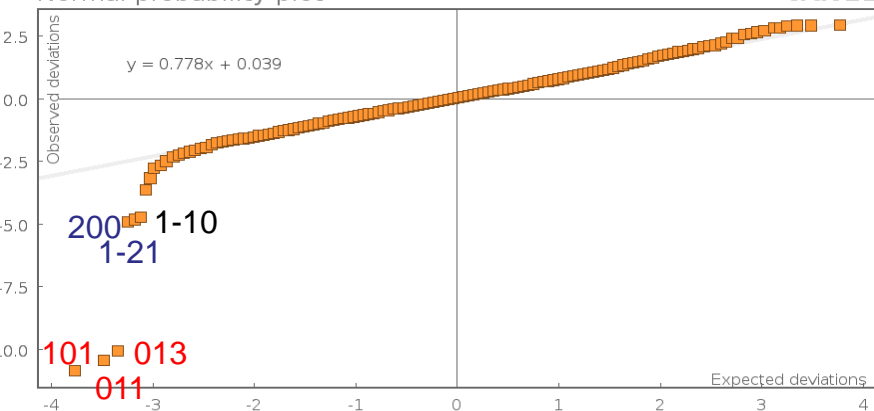
Decent structure, but three strongly disagreeable reflections.  $F_{\text{obs}} - F_{\text{calc}}$  plot shows that the most intense reflections are suppressed, which might indicate [extinction problems](#) or [detector saturation](#) problems. The normal probability plot is not symmetric. In cases which a few, clearly outlying reflections which share a common characteristic, we can suppress them using the OMIT command.



Fobs vs Fcalc



Normal probability plot



Most Disagreeable Reflections (\* if suppressed or used for Rfree).

h	k	l	Fo^2	Fc^2	Error/esd	Fc/Fc(max)	Res. (A)
1	0	1	33576.17	59459.27	10.52	0.853	8.36
0	1	1	37131.57	64288.11	10.13	0.886	8.14
0	1	3	48300.14	81807.84	9.75	1.000	4.70
2	0	0	38854.66	50922.42	4.91	0.789	4.85
1	-2	1	52253.97	73802.34	4.79	0.950	4.43
-1	1	0	24758.15	31810.84	4.73	0.624	7.12
-1	2	4	64.12	19.68	4.67	0.016	2.96

Home Work View Tools Info

Recent Files

Electron Density Peaks

Refinement Indicators

Bad Reflections

OMIT all reflections where |Error/esd| > than 5 Clear omits

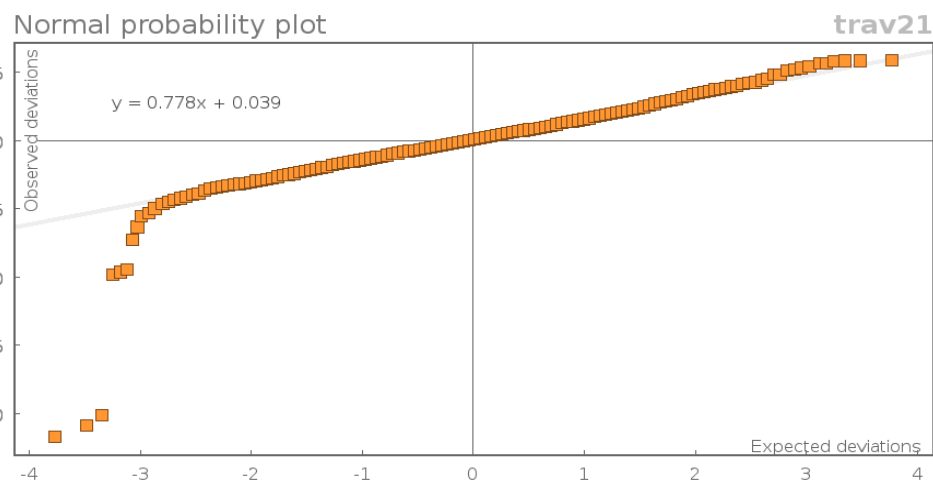
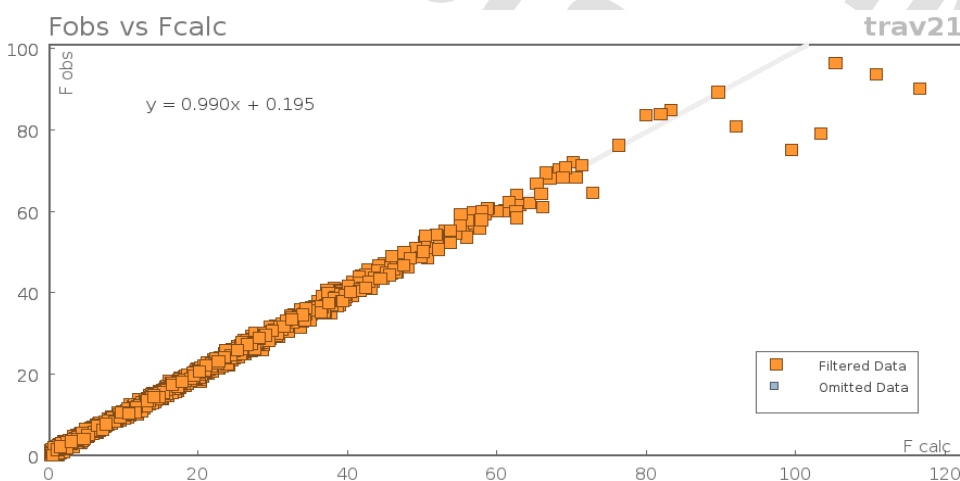
Unsigned sorting by Error/esd (refine to update)

H	K	L	Error/esd	omit	Edit...
1	0	1	10.52	omit	Edit...
0	1	1	10.13	omit	Edit...
0	1	3	9.75	omit	Edit...
2	0	0	4.91	omit	Edit...
1	-2	1	4.79	omit	Edit...
-1	1	0	4.73	omit	Edit...
-1	2	4	-4.67	omit	Edit...
3	0	9	-4.30	omit	Edit...
3	-1	8	-4.12	omit	Edit...
4	1	3	-4.04	omit	Edit...
-5	-2	1	-3.99	omit	Edit...
2	2	6	-3.96	omit	Edit...

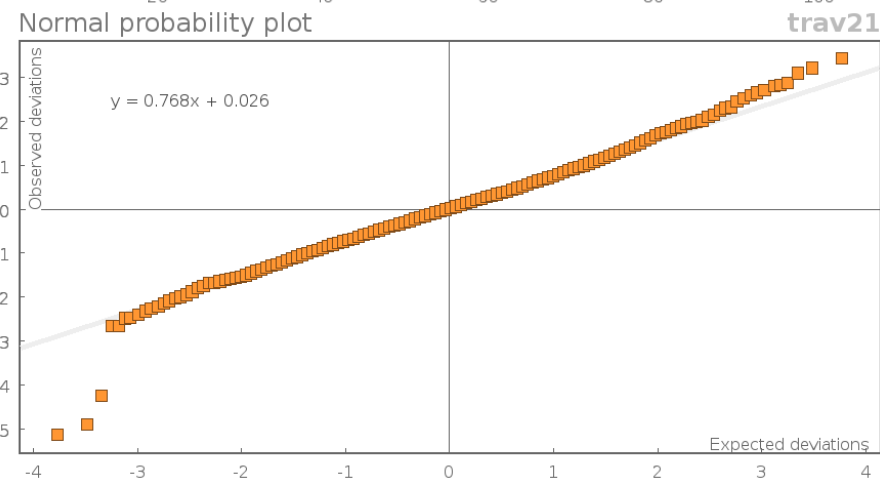
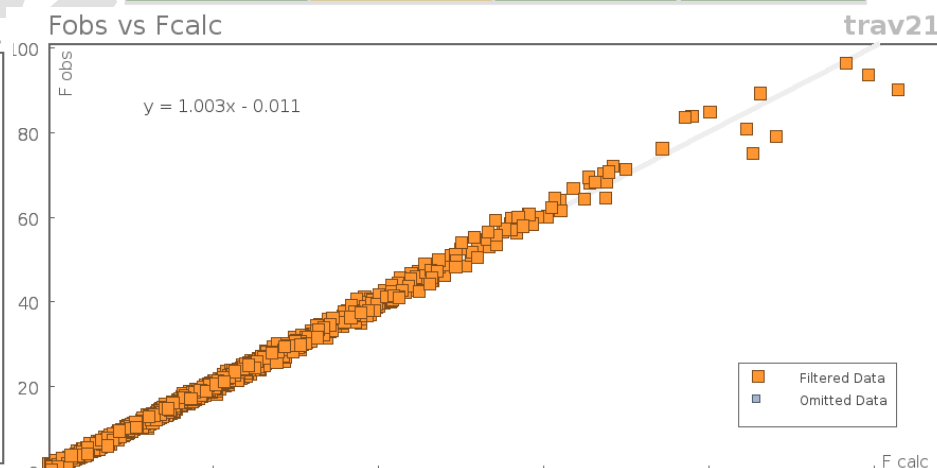


In this case, however, simply adding an EXTI command for extinction correction already reduces the problem.

## without EXTI

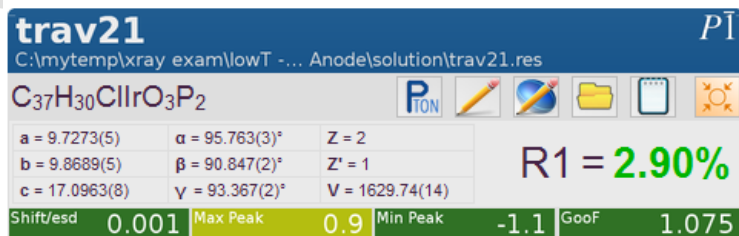


## with EXTI

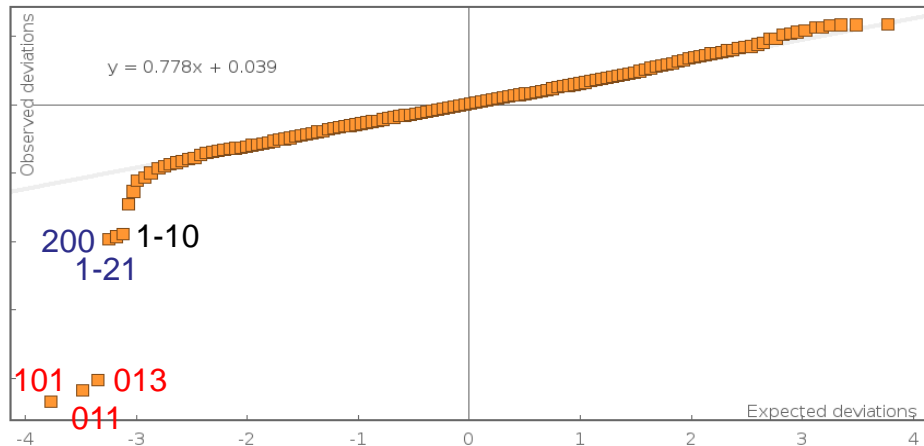
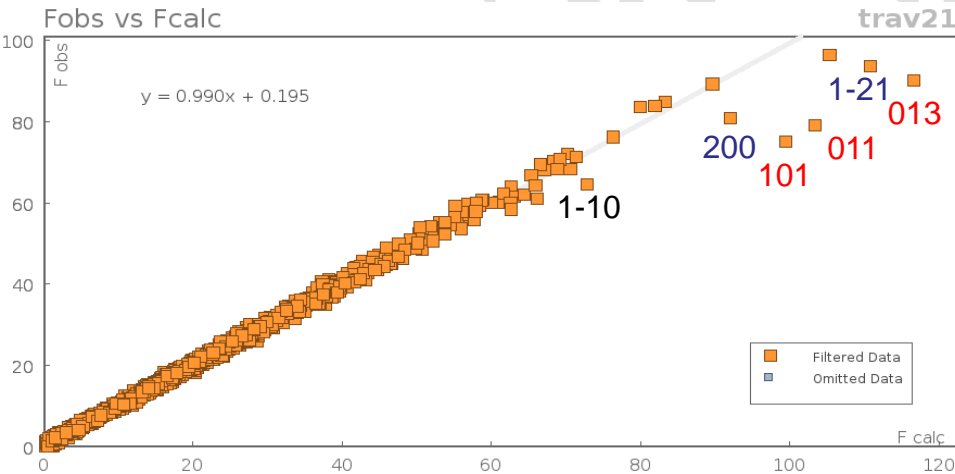


If EXTI would not have helped, we could suppress the worst reflections with OMIT.

without OMIT



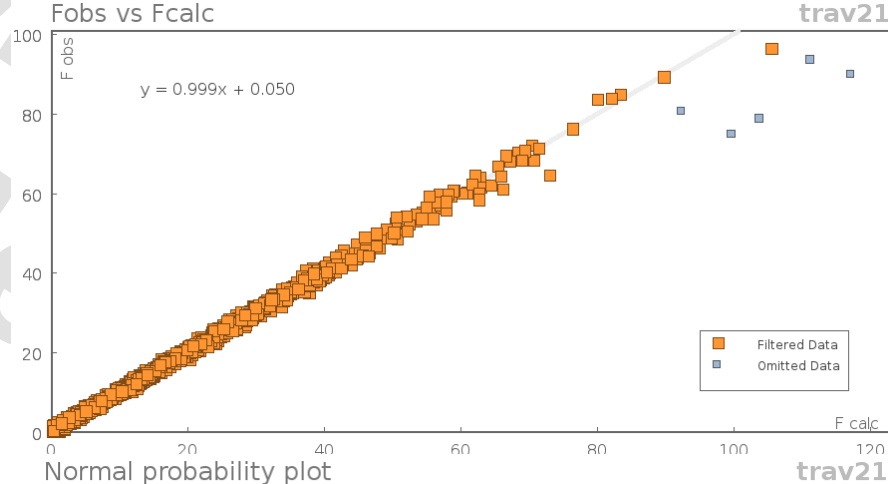
Fobs vs Fcalc



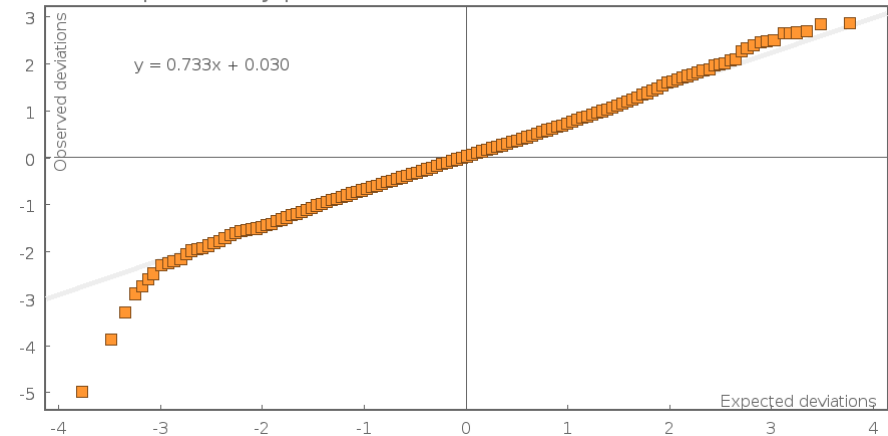
with OMIT

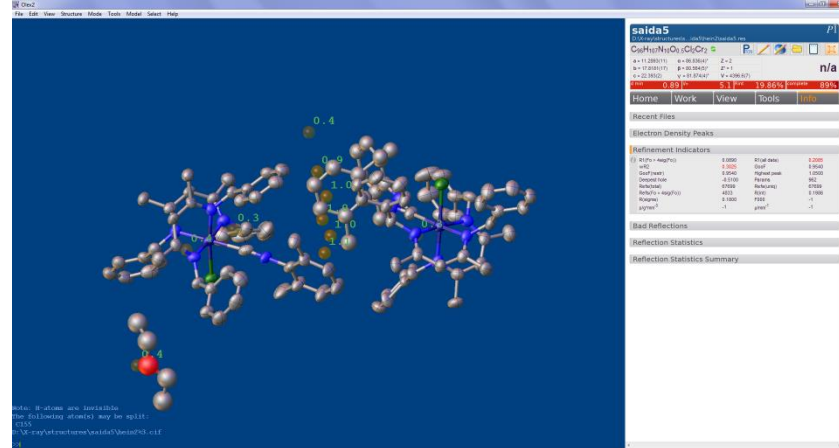


Fobs vs Fcalc

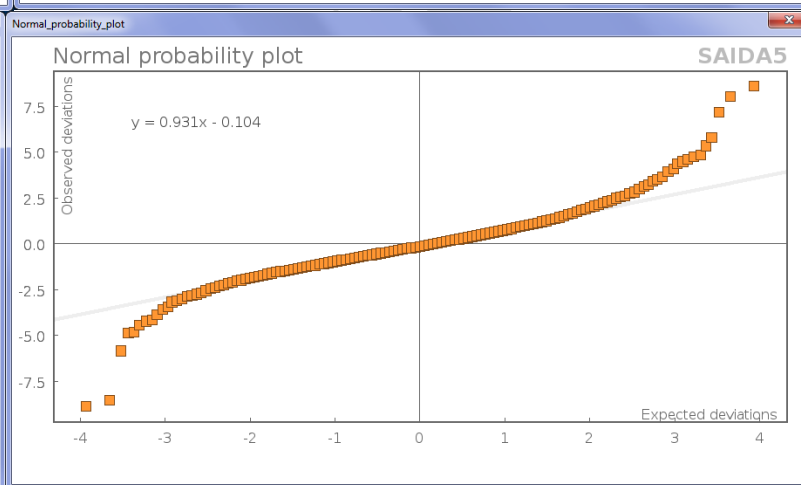
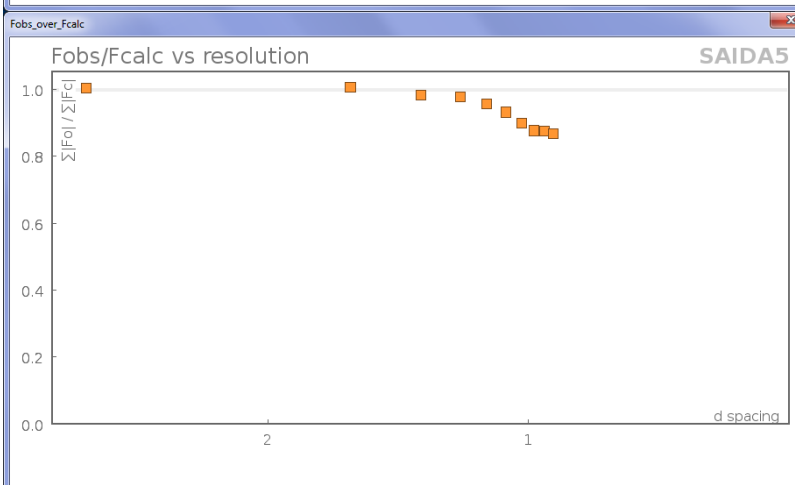
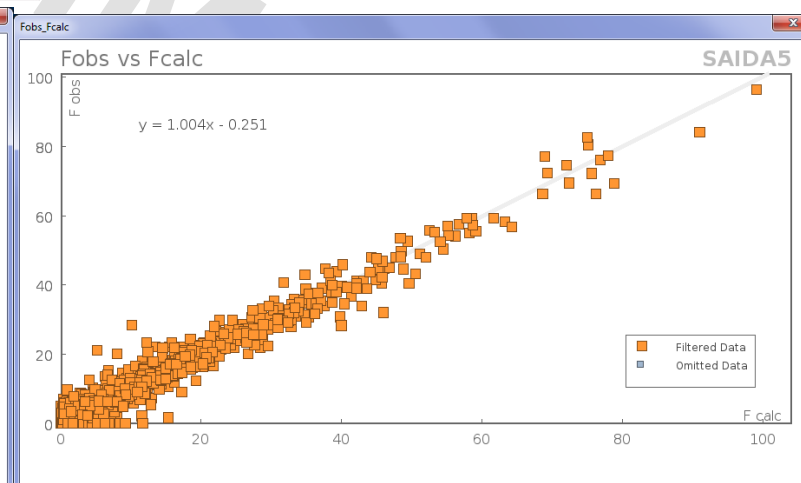
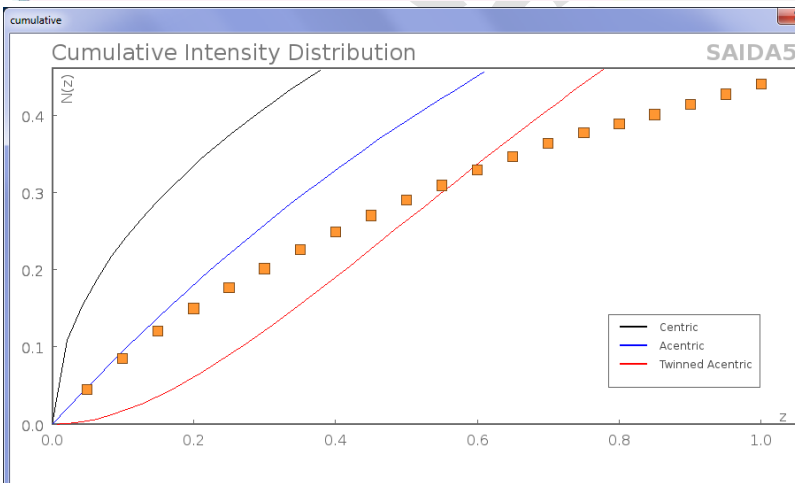


Normal probability plot



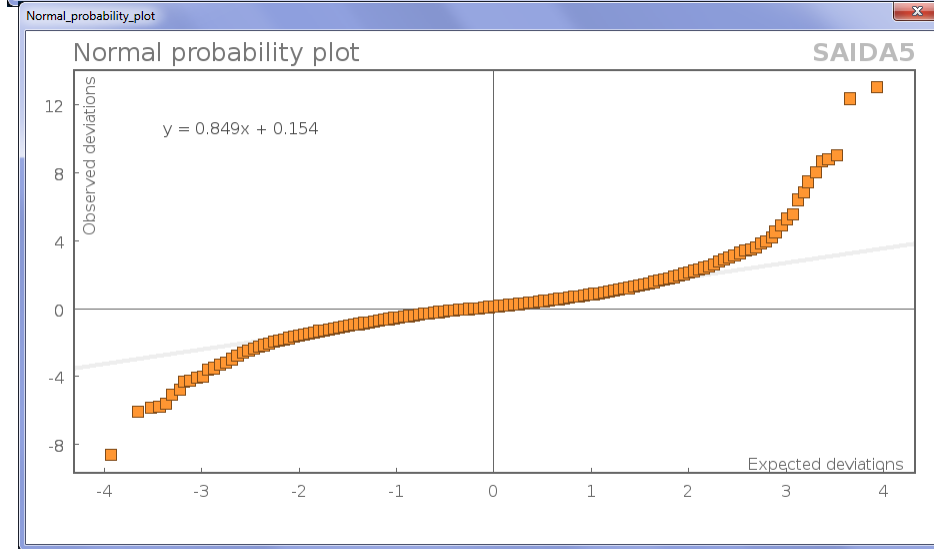
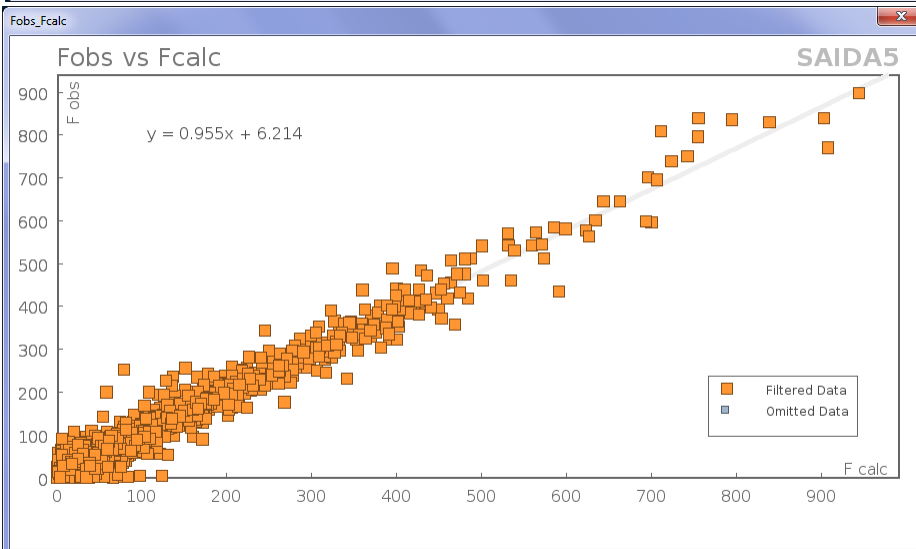
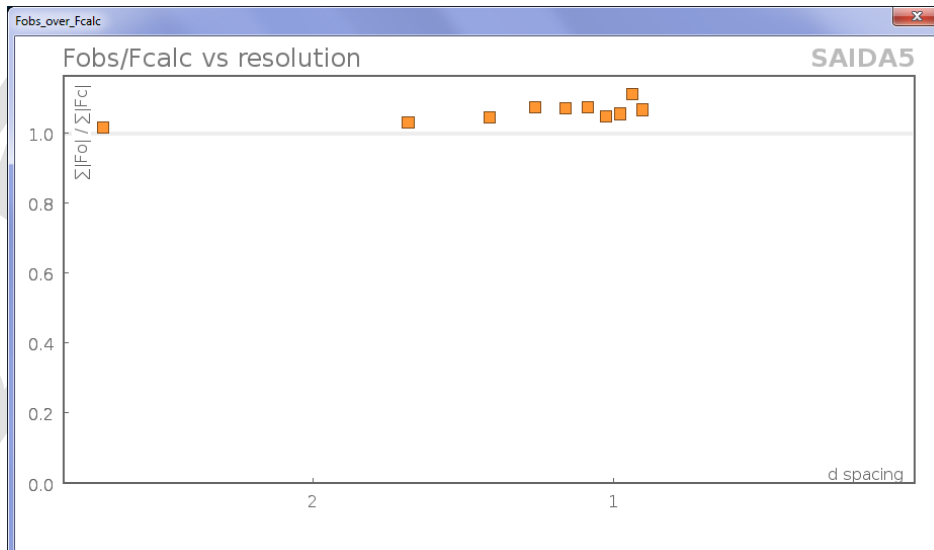
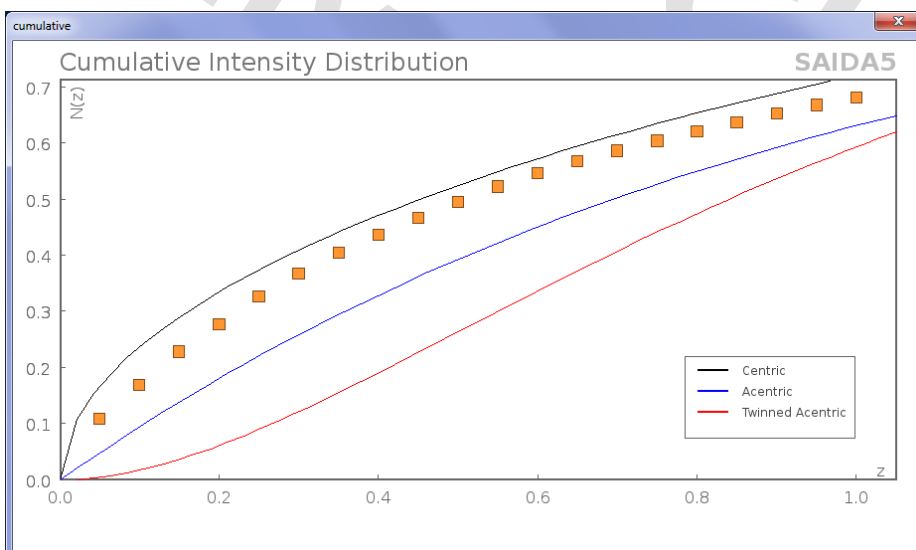


In case of strong disorder, the Cumulative Intensity plot might be depressed and Fobs/Fcalc decreases at higher resolution. (At high resolution disordered atoms show complete destructive interference.) Note that there is no notable influence on the probability plot.



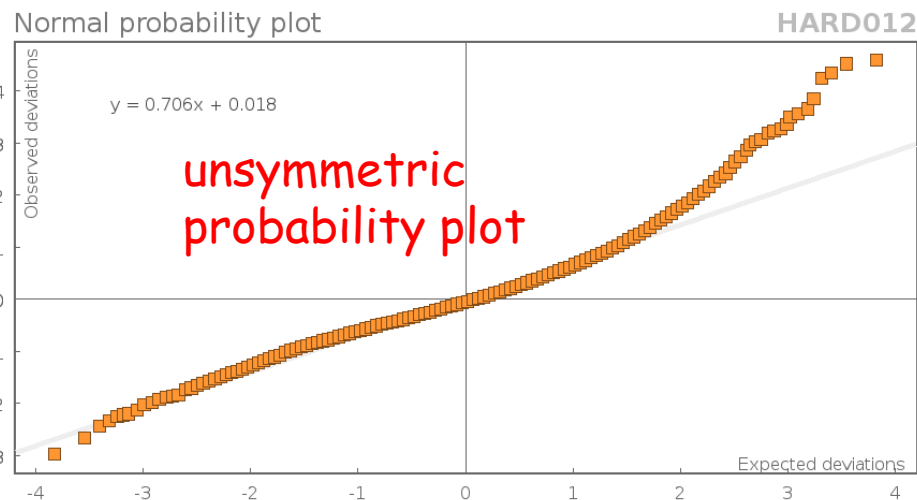
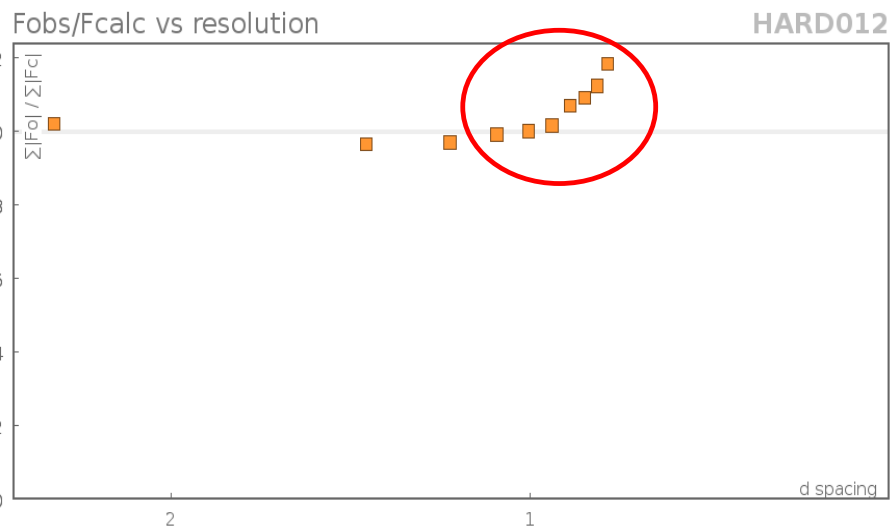
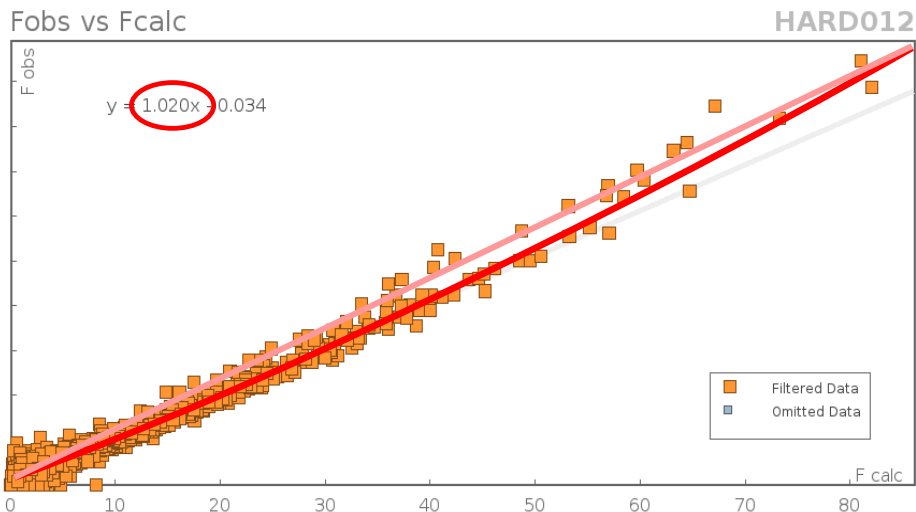
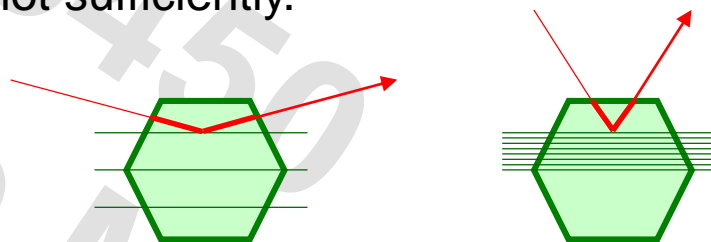
After application of SQUEEZE, the disorder effects on Cumulative Intensity and Fobs/Fcalc mostly vanished

Attention: this was a demonstration on the effects of strong disorder. **You should not use these plots as a justification to use SQUEEZE.**



Clearly something fishy is going on here.  $F_{\text{obs}}$  vs.  $F_{\text{calc}}$  is curved with *increasing* intensity at higher resolution. The probability plot is unsymmetric. The reason for this behaviour is not clear (yet), but clearly the dataset has problems.

One explanation might be strong absorption (this was a copper complex on Ga). At higher  $\theta$  angles the pathway in the crystal is shorter and absorption is lower. SADABS should have corrected for this, but eventually not sufficiently.



## **Bottom line:**

- Take the habit to check the statistics plot even for problem-free structures to establish your personal baseline of “How they should look like”.
- Not every deviation indicates a problem.
- If the R-value remains inexplicably high and you see problem indications in the statistical plot, you might want to verify your integration, absorption correction etc.