

Electron density is complex !

Göttingen, November 13th 2008

George M. Sheldrick, *Göttingen University*

<http://shelx.uni-ac.gwdg.de/SHELX/>

Friedel's Law

$$|F_{h,k,\ell}| = |F_{-h,-k,-\ell}| \quad \text{and} \quad \phi_{h,k,\ell} = -\phi_{-h,-k,-\ell}$$

Friedel's law holds exactly if there is no anomalous scattering. It can also be expressed in the form:

$$F_{h,k,\ell} = F_{-h,-k,-\ell}^*$$

where F^* is the complex conjugate of F . When Friedel's law holds, the structure factors are described by an *Hermitian* matrix and their Fourier transform (the electron density ρ) is, according to theory, *real*:

$$\rho_{x,y,z} = \sum_{h,k,\ell} F_{h,k,\ell} \exp[-2\pi i (hx+ky+\ell z)]$$

This summation includes all reflections, i.e. $-h,-k,-\ell$ as well as h,k,ℓ .

Complex scattering factors

The scattering factor of an atom is normally divided into three components:

$$f = f_0 + f' + i f''$$

For X-ray diffraction, f_0 depends on the resolution but not the wavelength; at $\sin(\theta)/\lambda = 0$ it is equal to the atomic number. The real component f' and the imaginary component f'' arise from resonance with electronic transitions in the atom, and are wavelength but not resolution dependent; both are usually relatively small.

A logical conclusion would be that the electron density should also be mainly real but with small and rather sharp imaginary components at the atomic positions, even for centrosymmetric space groups!

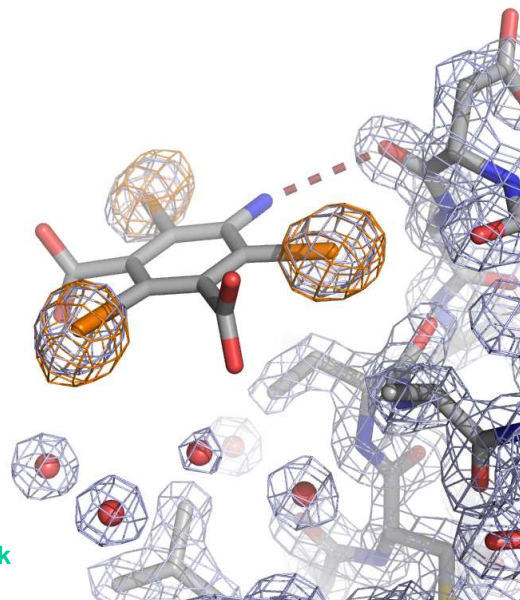
Anisotropy of the anomalous scattering

The f' part of the atomic scattering factor is in fact anisotropic. The magnitude of the scattering depends on the degree of polarization of the incident beam and the orientation of polarization direction relative to the crystal. Synchrotrons beams, in contrast to laboratory sources, have a high degree of polarization. This could have a marked effect on the imaginary part of an electron density map determined with synchrotron data, and can also cause systematically absent reflections to have significant intensity.

See many papers by Templeton & Templeton, also Schiltz & Bricogne, *Acta Cryst. D64* (2008) 711-729.

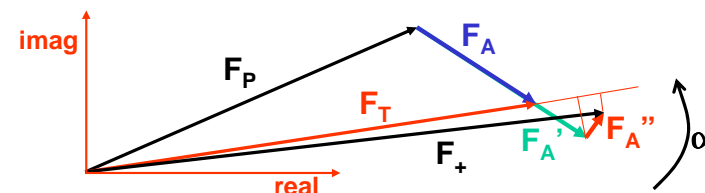
Macromolecular applications

Anomalous scattering is used for experimental phasing of macromolecules by the SAD and MAD methods. Frequently the 'real' density is contoured in blue and the 'imaginary density' in orange. The three iodine atoms on the ligand in this picture have both real and anomalous density, but the remaining atoms have only real density.



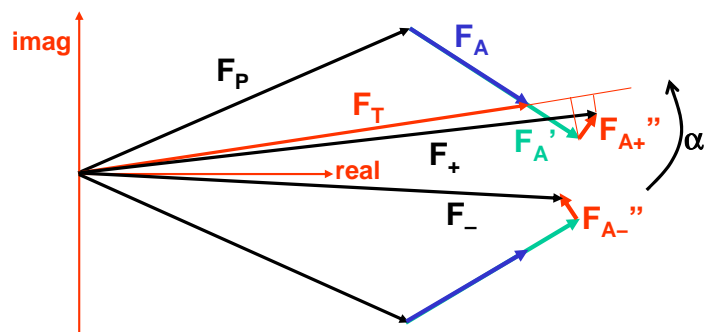
Structure factor vector diagram

Assuming that all the anomalous atoms are of the same element, we can divide the total non-anomalous structure factor F_T into the normal structure factor for the anomalous atoms, F_A , and the structure factor for the rest of the structure, F_P . To get the total structure factor in the presence of anomalous scattering we then need to add the real f' contribution F_A' and the imaginary f'' contribution F_A'' :



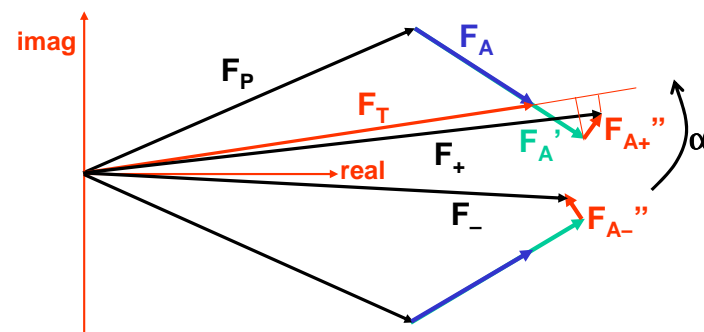
Where $\alpha = \phi_T - \phi_A$ is the phase shift that should be added to ϕ_A (the phase of F_A) to give the phase of F_T . Note that f' is normally negative, so F_A' is parallel to $-F_A$ rather than to $+F_A$ as shown here.

Friedel's law does not hold if f'' is not zero



When F_- is added to the diagram it becomes clear that if f'' is non-zero for at least one atom, and hence F_A'' is not zero, $|F_+| \neq |F_-|$ and so Friedel's law is violated. This turns out to be very useful, e.g. for the determination of *absolute configuration* or for *SAD phasing of macromolecules*.

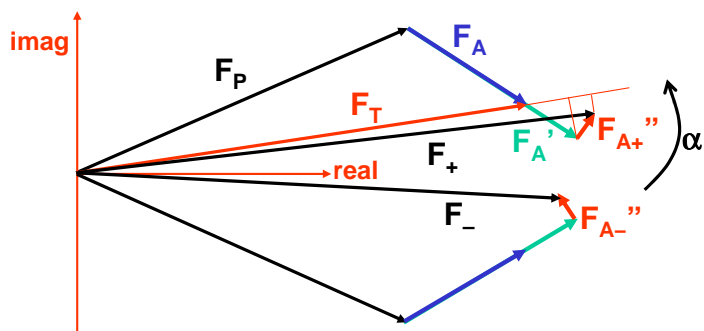
SAD phasing of macromolecules



If $|F_+| \gg |F_-|$, F_{A+}'' will be aligned close to $+F_+$ but F_{A-}'' will be pointing along $-F_-$. The angle α must then be close to $+90^\circ$. If $|F_-| \gg |F_+|$, α will be close to -90° . Both cases give a strong SAD phase indication.

However if the $|F_+| - |F_-|$ is close to zero, either F_{A+}'' and F_{A-}'' are very small or they must be approximately perpendicular to F_+ and F_- respectively, which is consistent with either α close to 0° or α close to 180° , a *twofold phase ambiguity*.

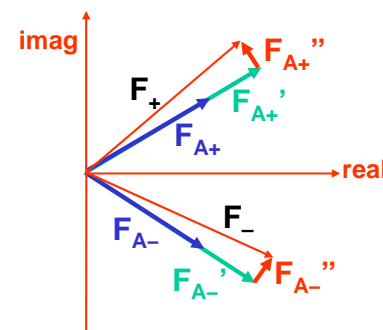
Anomalous (+90°) Fourier maps



Instead of $\phi_T \approx \phi_A + \alpha$ as in SAD phasing, if we know ϕ_T (because we have already solved the structure), we can find the anomalous atoms from a Fourier map with amplitudes $\Delta F = |F_+| - |F_-|$ and phases $\phi_T - 90^\circ$ (which is the same as amplitudes $|\Delta F|$ and phases $\phi_T - \alpha \approx \phi_A$). This is a good approximation when $|\Delta F|$ is large, The small $|\Delta F|$ values with unreliable phases just add a little noise to the map (Kraut (1968) *JMB* 35, 511-512; http://skuld.bmsc.washington.edu/scatter/AS_Bijvoet.html).

Special case: α -Se

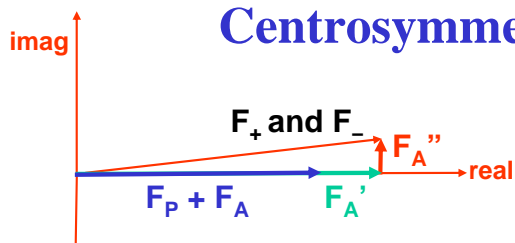
The α modification of elemental selenium crystallizes in $P3_221$ or $P3_121$ with infinite helices along the 3_1 or 3_2 axes. Since no atoms of other elements are present, F_P is zero so $F_T = F_A$.



As can be seen from the diagram, $|F_+|$ and $|F_-|$ are equal in such a case, so they cannot be used to determine the chirality of the helices in a given crystal!

Only the first part of Friedel's law [$|F_{h,k,\ell}| = |F_{-h,-k,-\ell}|$] holds, not the second part [$\phi_{h,k,\ell} = -\phi_{-h,-k,-\ell}$]!

Centrosymmetric structures



In this case also, only the first part of Friedel's law [$|F_{h,k,\ell}| = |F_{-h,-k,-\ell}|$] holds, not the second part [$\phi_{h,k,\ell} = -\phi_{-h,-k,-\ell}$]. The contributions to F_+ and F_- are identical and the phase angles of $\phi_{h,k,\ell}$ and $\phi_{-h,-k,-\ell}$ are the same but not 0 or 180°. So the structure factors are not Hermitian and a Fourier synthesis should give complex electron density (were it not for the fact that all programs assume $\phi_{h,k,\ell} = -\phi_{-h,-k,-\ell}$).

Since we cannot determine the phase angles, the imaginary part of the electron density is *experimentally inaccessible* for centrosymmetric structures, and can only be determined *approximately* for non-centrosymmetric structures. Reflections for which F_{A+}' is not zero but $F_P + F_A + F_{A+}'$ accidentally cancels to zero might however lead to errors in maps; SHELXL subtracts the anomalous contribution from $|F_o|$ and $|F_c|$ before writing them to the .fcf file to minimize this error.

A charge density study in $P2_1/n$

If only the refinement and Fourier programs would not assume $\phi_{h,k,\ell} = -\phi_{-h,-k,-\ell}$ the results might look like:

h	k	ℓ	$ F_{A+}' $	$ F_{\pm} $	ϕ
-21	4	1	0.32	4.20	4.4
21	-4	-1	0.32	4.20	4.4
-20	4	1	0.37	0.49	50.1
20	-4	-1	0.37	0.49	50.1
-19	4	1	1.18	18.59	3.7
19	-4	-1	1.18	18.59	3.7
-18	4	1	0.99	1.05	-69.8
18	-4	-1	0.99	1.05	-69.8
-17	4	1	0.56	25.55	-178.7
17	-4	-1	0.56	25.55	-178.7
-16	4	1	0.83	22.05	-177.8
16	-4	-1	0.83	22.05	-177.8
-15	4	1	1.11	9.21	-7.0
15	-4	-1	1.11	9.21	-7.0
-14	4	1	1.80	44.35	2.3
14	-4	-1	1.80	44.35	2.3
-13	4	1	0.21	11.54	178.9
13	-4	-1	0.21	11.54	178.9

