2.3. Patterson and molecular replacement techniques, and the use of noncrystallographic symmetry in phasing

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2.3.1. Introduction

2.3.1.1. Background

Historically, the Patterson has been used in a variety of ways to effect the solutions of crystal structures. While some simple structures (Ketelaar & de Vries, 1939; Hughes, 1940; Speakman, 1949; Shoemaker *et al.*, 1950) were solved by direct analysis of Patterson syntheses, alternative methods have largely superseded this procedure. An early innovation was the heavy-atom method which depends on the location of a small number of relatively strong scatterers (Harker, 1936). Image-seeking methods and Patterson superposition techniques were first contemplated in the late 1930s (Wrinch, 1939) and applied sometime later (Beevers & Robertson, 1950; Clastre & Gay, 1950; Garrido, 1950*a*; Buerger, 1959). This experience provided the encouragement for computerized vector-search methods to locate individual atoms automatically (Mighell & Jacobson, 1963; Kraut, 1961; Hamilton, 1965; Simpson *et al.*, 1965) or to position known molecular fragments in unknown crystal structures (Nordman & Nakatsu, 1963; Huber, 1965). The Patterson function has been used extensively in conjunction with the isomorphous replacement method (Rossmann, 1960; Blow, 1958) or anomalous dispersion (Rossmann, 1961*a*) to determine the position of heavy-atom substitution. Pattersons have been used to detect the presence and relative orientation of multiple copies of a given chemical motif in the crystallographic asymmetric unit in the same or different crystals (Rossmann & Blow, 1962). Finally, the orientation and placement of known molecular structures ('molecular replacement') into unknown crystal structures can be accomplished *via* Patterson techniques.

The function, introduced by Patterson in 1934 (Patterson, 1934*a*,*b*), is a convolution of electron density with itself and may be defined as

$$P(\mathbf{u}) = \int_V \rho(\mathbf{x}) \cdot \rho(\mathbf{u} + \mathbf{x}) \, d\mathbf{x}, \qquad (2.3.1.1)$$

where $P(\mathbf{u})$ is the 'Patterson' function at $\mathbf{u}$, $\rho(\mathbf{x})$ is the crystal's periodic electron density and V is the volume of the unit cell. The Patterson function, or F^2 series, can be calculated directly from the experimentally derived X-ray intensities as

$$P(\mathbf{u}) = \frac{2}{V^2} \sum_{\mathbf{h}}^{\text{hemisphere}} |\mathbf{F_h}|^2 \cos 2\pi \mathbf{h} \cdot \mathbf{u}. \qquad (2.3.1.2)$$

The derivation of (2.3.1.2) from (2.3.1.1) can be found in this volume (see Section 1.3.4.2.1.6) along with a discussion of the physical significance and symmetry of the Patterson function, although the principal properties will be restated here.

The Patterson can be considered to be a vector map of all the pairwise interactions between the atoms in a unit cell. The vectors in a Patterson correspond to vectors in the real (direct) crystal cell but translated to the Patterson origin. Their weights are proportional to the product of densities at the tips of the vectors in the real cell. The Patterson unit cell has the same size as the real crystal cell. The symmetry of the Patterson comprises the Laue point group of the crystal cell plus any additional lattice symmetry due to Bravais centring. The reduction of the real

space group to the Laue symmetry is produced by the translation of all vectors to the Patterson origin and the introduction of a centre of symmetry. The latter is a consequence of the relationship between the vectors **AB** and **BA**. The Patterson symmetries for all 230 space groups are tabulated in *IT* A (2005).

An analysis of Patterson peaks can be obtained by considering N atoms with form factors f_i in the unit cell. Then

$$\mathbf{F_h} = \sum_{i=1}^{N} f_i \exp(2\pi i \mathbf{h} \cdot \mathbf{x}_i).$$

Using Friedel's law,

$$|\mathbf{F_h}|^2 = \mathbf{F_h} \cdot \mathbf{F_h^*}$$
$$= \left[\sum_{i=1}^{N} f_i \exp(2\pi i \mathbf{h} \cdot \mathbf{x}_i) \right] \left[\sum_{j=1}^{N} f_j \exp(-2\pi i \mathbf{h} \cdot \mathbf{x}_j) \right],$$

which can be decomposed to

$$|\mathbf{F_h}|^2 = \sum_{i=1}^{N} f_i^2 + \sum_{i \neq j}^{N} \sum^{N} f_i f_j \exp[2\pi i \mathbf{h} \cdot (\mathbf{x}_i - \mathbf{x}_j)]. \qquad (2.3.1.3)$$

On substituting (2.3.1.3) in (2.3.1.2), we see that the Patterson consists of the sum of N^2 total interactions of which N are of weight f_i^2 at the origin and $N(N-1)$ are of weight $f_i f_j$ at $\mathbf{x}_i - \mathbf{x}_j$.

The weight of a peak in a real cell is given by

$$w_i = \int_U \rho_i(\mathbf{x}) \, d\mathbf{x} = Z_i \text{ (the atomic number)},$$

where U is the volume of the atom i. By analogy, the weight of a peak in a Patterson (form factor $f_i f_j$) will be given by

$$w_{ij} = \int_U P_{ij}(\mathbf{u}) \, d\mathbf{u} = Z_i Z_j.$$

Although the maximum height of a peak will depend on the spread of the peak, it is reasonable to assume that heights of peaks in a Patterson are proportional to the products of the atomic numbers of the interacting atoms.

There are a total of N^2 interactions in a Patterson due to N atoms in the crystal cell. These can be represented as an $N \times N$ square matrix whose elements $\mathbf{u}_{ij}$, w_{ij} indicate the position and weight of the peak produced between atoms i and j (Table 2.3.1.1). The N vectors corresponding to the diagonal of this matrix are located at the Patterson origin and arise from the convolution of each atom with itself. This leaves $N(N-1)$ vectors whose locations depend on the relative positions of all of the atoms in the crystal cell and whose weights depend on the atom types related by the vector. Complete specification of the unique non-origin Patterson vectors requires description of only the $N(N-1)/2$ elements in either the upper or the lower triangle of this matrix, since the two sets of vectors represented by the two triangles are related by a centre of symmetry

Table 2.3.1.1. *Matrix representation of Patterson peaks*

The $N \times N$ matrix represents the position $\mathbf{u}_{ij}$ and weights w_{ij} of atomic interactions in a Patterson arising from N atoms at $\mathbf{x}_i$ and weight w_i in the real cell.

	$\mathbf{x}_1, w_1$	$\mathbf{x}_2, w_2$	$\ldots$	$\mathbf{x}_N, w_N$
$\mathbf{x}_1, w_1$	$\mathbf{u}_{11} = \mathbf{x}_1 - \mathbf{x}_1,$ $w_{11} = w_1^2$	$\mathbf{u}_{12} = \mathbf{x}_1 - \mathbf{x}_2,$ $w_{12} = w_1 w_2$	$\ldots$	$\mathbf{u}_{1N} = \mathbf{x}_1 - \mathbf{x}_N,$ $w_{1N} = w_1 w_N$
$\mathbf{x}_2, w_2$	$\mathbf{x}_2 - \mathbf{x}_1, w_2 w_1$	$0, w_2^2$	$\ldots$	$\mathbf{x}_2 - \mathbf{x}_N, w_2 w_N$
$\vdots$	$\vdots$	$\vdots$	$\ddots$	$\vdots$
$\mathbf{x}_N, w_N$	$\mathbf{x}_N - \mathbf{x}_1, w_N w_1$	$\mathbf{x}_N - \mathbf{x}_2, w_N w_2$	$\ldots$	$0, w_N^2$

$[\mathbf{u}_{ij} \equiv \mathbf{x}_i - \mathbf{x}_j = -\mathbf{u}_{ij} \equiv -(\mathbf{x}_j - \mathbf{x}_i)]$. Patterson vector positions are usually represented as $\langle uvw \rangle$, where u, v and w are expressed as fractions of the Patterson cell axes.

2.3.1.2. Limits to the number of resolved vectors

If we assume a constant number of atoms per unit volume, the number of atoms N in a unit cell increases in direct proportion with the volume of the unit cell. Since the number of non-origin peaks in the Patterson function is $N(N-1)$ and the Patterson cell is the same size as the real cell, the problem of overlapping peaks in the Patterson function becomes severe as N increases. To make matters worse, the breadth of a Patterson peak is roughly equal to the sum of the breadth of the original atoms. The effective width of a Patterson peak will also increase with increasing thermal motion, although this effect can be artificially reduced by sharpening techniques. Naturally, a loss of attainable resolution at high scattering angles will affect the resolution of atomic peaks in the real cell as well as peaks in the Patterson cell. If U is the van der Waals volume per average atom, then the fraction of the cell occupied by atoms will be $f = NU/V$. Similarly, the fraction of the cell occupied by Patterson peaks will be $2UN(N-1)/V$ or $2f(N-1)$. With the reasonable assumption that $f \simeq 0.1$ for a typical organic crystal, then the cell can contain at most five atoms ($N \leq 5$) for there to be no overlap, other than by coincidence, of the peaks in the Patterson. As N increases there will occur a background of peaks on which are superimposed features related to systematic properties of the structure.

The contrast of selected Patterson peaks relative to the general background level can be enhanced by a variety of techniques. For instance, the presence of heavy atoms not only enhances the size of a relatively small number of peaks but ordinarily ensures a larger separation of the peaks due to the light-atom skeleton on which the heavy atoms are hung. That is, the factor f (above) is substantially reduced. Another example is the effect of systematic atomic arrangements (*e.g.* α-helices or aromatic rings) resulting in multiple peaks which stand out above the background. In the isomorphous replacement method, isomorphous difference Pattersons are utilized in which the contrast of the Patterson interactions between the heavy atoms is enhanced by removal of the predominant interactions which involve the rest of the structure.

2.3.1.3. Modifications: origin removal, sharpening etc.

A. L. Patterson, in his first in-depth exposition of his newly discovered F^2 series (Patterson, 1935), introduced the major modifications to the Patterson which are still in use today. He illustrated, with one-dimensional Fourier series, the techniques of removing the Patterson origin peak, sharpening the overall function and also removing peaks due to atoms in special positions. Each one of these modifications can improve the interpretability of Pattersons, especially those of simple structures. Whereas the recommended extent of such modifications is controversial (Buerger, 1966), most studies which utilize Patterson functions do incorporate some of these techniques [see,

for example, Jacobson *et al.* (1961), Braun *et al.* (1969) and Nordman (1980*a*)]. Since Patterson's original work, other workers have suggested that the Patterson function itself might be modified; Fourier inversion of the modified Patterson then provides a new and perhaps more tractable set of structure factors (McLachlan & Harker, 1951; Simonov, 1965; Raman, 1966; Corfield & Rosenstein, 1966). Karle & Hauptman (1964) suggested that an improved set of structure factors could be obtained from an origin-removed Patterson modified such that it was everywhere non-negative and that Patterson density values less than a bonding distance from the origin were set to zero. Nixon (1978) was successful in solving a structure which had previously resisted solution by using a set of structure factors which had been obtained from a Patterson in which the largest peaks had been attenuated.

The N origin peaks [see expression (2.3.1.3)] may be removed from the Patterson by using coefficients

$$|\mathbf{F}_{\mathbf{h}, \, \text{mod}}|^2 = |\mathbf{F}_{\mathbf{h}}|^2 - \sum_{i=1}^{N} f_i^2.$$

A Patterson function using these modified coefficients will retain all interatomic vectors. However, the observed structure factors $\mathbf{F}_{\mathbf{h}}$ must first be placed on an absolute scale (Wilson, 1942) in order to match the scattering-factor term. In practice, Patterson origins can also be removed by using coefficients

$$|\mathbf{F}_{\mathbf{h},\text{mod}}|^2 = |\mathbf{F}_{\mathbf{h}}|^2 - \langle |\mathbf{F}_{\mathbf{h}}|^2 \rangle,$$

where $\langle |\mathbf{F}_{\mathbf{h}}|^2 \rangle$ is the average reflection intensity, usually calculated in several resolution shells. This formula has the advantage that the observed structure factors do not need to be on absolute scale.

Analogous to origin removal, the vector interactions due to atoms in known positions can also be removed from the Patterson function. Patterson showed that non-origin Patterson peaks arising from known atoms 1 and 2 may be removed by using the expression

$$|\mathbf{F}_{\mathbf{h}, \, \text{mod}}|^2 = |\mathbf{F}_{\mathbf{h}}|^2 - \sum_{i=1}^{N} f_i^2 t_i^2 - 2 f_1 f_2 t_1 t_2 \cos 2\pi \mathbf{h} \cdot (\mathbf{x}_1 - \mathbf{x}_2),$$

where $\mathbf{x}_1$ and $\mathbf{x}_2$ are the positions of atoms 1 and 2 and t_1 and t_2 are their respective thermal correction factors. Using one-dimensional Fourier series, Patterson illustrated how interactions due to known atoms can obscure other information.

Patterson also introduced a means by which the peaks in a Patterson function may be artificially sharpened. He considered the effect of thermal motion on the broadening of electron-density peaks and consequently their Patterson peaks. He suggested that the F^2 coefficients could be corrected for thermal effects by simulating the atoms as point scatterers and proposed using a modified set of coefficients

$$|\mathbf{F}_{\mathbf{h}, \, \text{sharp}}|^2 = |\mathbf{F}_{\mathbf{h}}|^2 / \bar{f}^2,$$

where $\bar{f}$, the average scattering factor per electron, is given by

$$\bar{f} = \sum_{i=1}^{N} f_i \bigg/ \sum_{i=1}^{N} Z_i.$$

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