2.3. PATTERSON AND MOLECULAR REPLACEMENT TECHNIQUES
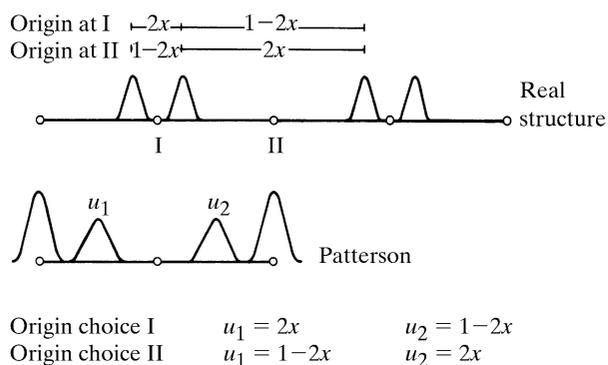

Fig. 2.3.2.1. Origin selection in the interpretation of a Patterson of a one-dimensional centrosymmetric structure.

Fig. 2.3.2.2. The *c*-axis projection of cuprous chloride azomethane complex $(C_2H_6Cl_2Cu_2N_2)$. The space group is $P\bar{1}$ with one molecule per unit cell. [Adapted from and reprinted with permission from Woolfson (1970, p. 321).]

means for describing homometric structures [Buerger (1959, pp. 41–50), Menzer (1949), Bullough (1961, 1964), Hoppe (1962)].

Since a Patterson function is centrosymmetric, the Pattersons of a crystal structure and of its mirror image are identical. Thus the enantiomeric ambiguity present in noncentrosymmetric crystal structures cannot be overcome by using the Patterson alone and represents a special case of homometric structures. Assignment of the correct enantiomorph in a crystal structure analysis is generally not possible unless a recognizable fragment of known chirality emerges (*e.g.* L-amino acids in proteins, D-riboses in nucleic acids, the known framework of steroids and other natural products, the right-handed twist of α-helices, the left-handed twist of successive strands in a β-sheet, the fold of a known protein subunit *etc.*) or anomalous-scattering information is available and can be used to resolve the ambiguity (Bijvoet *et al.*, 1951).

It is frequently necessary to select arbitrarily one enantiomorph over another in the early stages of a structure solution. Structure-factor phases calculated from a single heavy atom in space group $P1$, $P2$ or $P2_1$ (for instance) will be centrosymmetric and both enantiomorphs will be present in Fourier calculations based on these phases. In other space groups (*e.g.* $P2_12_12_1$), the selected heavy atom is likely to be near one of the planes containing the 2_1 axes and thus produce a weaker 'ghost' image of the opposite enantiomorph. The mixture of the two overlapped enantiomorphic solutions can cause interpretive difficulties. As the structure solution progresses, the 'ghosts' are exorcized owing to the dominance of the chosen enantiomorph in the phases.

2.3.1.5. *The Patterson synthesis of the second kind*

Patterson also defined a second, less well known, function (Patterson, 1949) as

$$P_{\pm}(\mathbf{u}) = \int \rho(\mathbf{u}+\mathbf{x}) \cdot \rho(\mathbf{u}-\mathbf{x}) \, d\mathbf{x}$$

$$= \frac{2}{V^2} \sum_{\mathbf{h}}^{\text{hemisphere}} F_{\mathbf{h}}^2 \cos(2\pi 2\mathbf{h} \cdot \mathbf{u} - 2\alpha_{\mathbf{h}}).$$

This function can be computed directly only for centrosymmetric structures. It can be calculated for noncentrosymmetric structures when the phase angles are known or assumed. It will represent the degree to which the known or assumed structure has a centre of symmetry at $\mathbf{u}$. That is, the product of the density at $\mathbf{u}+\mathbf{x}$ and $\mathbf{u}-\mathbf{x}$ is large when integrated over all values $\mathbf{x}$ within the unit cell. Since atoms themselves have a centre of symmetry, the function will contain peaks at each atomic site roughly proportional in height to the square of the number of electrons in each atom plus peaks at the midpoint between atoms proportional in
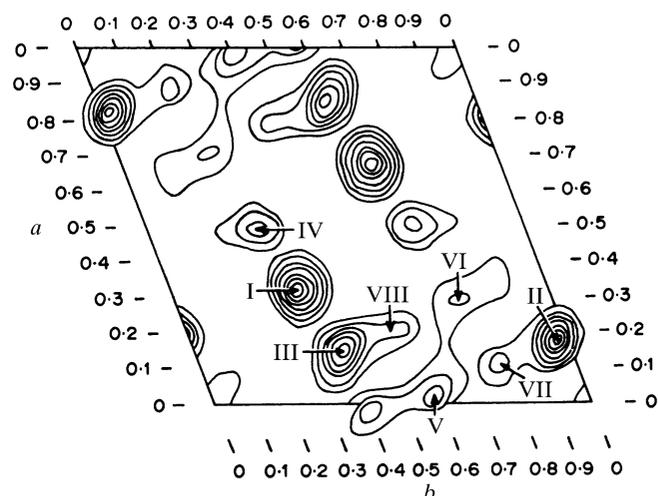
height to the product of the electrons in each atom. Although this function has not been found very useful in practice, it is useful for demonstrating the presence of weak enantiomorphic images in a given tentative structure determination.

2.3.2. Interpretation of Patterson maps

2.3.2.1. Simple solutions in the triclinic cell. Selection of the origin

A hypothetical one-dimensional centrosymmetric crystal structure containing an atom at x and at $-x$ and the corresponding Patterson is illustrated in Fig. 2.3.2.1. There are two different centres of symmetry which may be chosen as convenient origins. If the atoms are of equal weight, we expect Patterson vectors at positions $u = \pm 2x$ with weights equal to half the origin peak. There are two symmetry-related peaks, u_1 and u_2 (Fig. 2.3.2.1) in the Patterson. It is an arbitrary choice whether $u_1 = 2x$ or $u_2 = 2x$. This choice is equivalent to selecting the origin at the centre of symmetry I or II in the real structure (Fig. 2.3.2.1). Similarly in a three-dimensional $P\bar{1}$ cell, the Patterson will contain peaks at $\langle uvw \rangle$ which can be used to solve for the atom coordinates $\pm\langle 2x, 2y, 2z \rangle$. Solving for the same coordinates by starting from symmetric representations of the same vector will lead to alternate origin choices. For example, use of $\langle 1+u, 1+v, w \rangle$ will lead to translating the origin by $(+\frac{1}{2}, +\frac{1}{2}, 0)$ relative to the solution based on $\langle uvw \rangle$. There are eight distinct inversion centres in $P\bar{1}$, each one of which represents a valid origin choice. Although any choice of origin would be allowable, an inversion centre is convenient because then the structure factors are all real. Typically, one of the vector peaks closest to the Patterson origin is selected to start the solution, usually in the calculated asymmetric unit of the Patterson. Care must be exercised in selecting the same origin for all atomic positions by considering cross-vectors between atoms.

Examine, for example, the *c*-axis Patterson projection of a cuprous chloride azomethane complex $(C_2H_6Cl_2Cu_2N_2)$ in $P\bar{1}$ as shown in Fig. 2.3.2.2. The largest Patterson peaks should correspond to vectors arising from Cu $(Z = 29)$ and Cl $(Z = 17)$ atoms. There will be copper atoms at $\mathbf{x}_{Cu}(x_{Cu}, y_{Cu})$ and $-\mathbf{x}_{Cu}(-x_{Cu}, -y_{Cu})$ as well as chlorine atoms at analogous positions. The interaction matrix is

	$\mathbf{x}_{Cu}$, 29	$\mathbf{x}_{Cl}$, 17	$-\mathbf{x}_{Cu}$, 29	$-\mathbf{x}_{Cl}$, 17
$\mathbf{x}_{Cu}$, 29	0, 841	$\mathbf{x}_{Cu} - \mathbf{x}_{Cl}$, 493	$2\mathbf{x}_{Cu}$, 841	$\mathbf{x}_{Cu} + \mathbf{x}_{Cl}$, 493
$\mathbf{x}_{Cl}$, 17		0, 289	$\mathbf{x}_{Cl} + \mathbf{x}_{Cu}$, 493	$2\mathbf{x}_{Cl}$, 289
$-\mathbf{x}_{Cu}$, 29			0, 841	$\mathbf{x}_{Cu} - \mathbf{x}_{Cl}$, 493
$-\mathbf{x}_{Cl}$, 17				0, 289

Table 2.3.2.1. *Coordinates of Patterson peaks for $C_2H_6Cl_2Cu_2N_2$ projection*

Height	u	v	Number in diagram (Fig. 2.3.2.2)
7	0.33	0.34	I
7	0.18	0.97	II
6	0.16	0.40	III
3	0.49	0.29	IV
3	0.02	0.59	V
2	0.30	0.75	VI
2	0.12	0.79	VII

which shows that the Patterson should contain the following types of vectors:

Position	Weight	Multiplicity	Total weight
$2\mathbf{x}_{Cu}$	841	1	841
$2\mathbf{x}_{Cl}$	289	1	289
$\mathbf{x}_{Cu} - \mathbf{x}_{Cl}$	493	2	986
$\mathbf{x}_{Cu} + \mathbf{x}_{Cl}$	493	2	986

The coordinates of the largest Patterson peaks are given in Table 2.3.2.1 for an asymmetric half of the cell chosen to span $0 \rightarrow \frac{1}{2}$ in u and $0 \rightarrow 1$ in v. Since the three largest peaks are in the same ratio (7:7:6) as the three largest expected vector types (986:986:841), it is reasonable to assume that peak III corresponds to the copper–copper interaction at $2\mathbf{x}_{Cu}$. Hence, $x_{Cu} = 0.08$ and $y_{Cu} = 0.20$. Peaks I and II should be due to the double-weight Cu–Cl vectors at $\mathbf{x}_{Cu} - \mathbf{x}_{Cl}$ and $\mathbf{x}_{Cu} + \mathbf{x}_{Cl}$. Now suppose that peak I is at position $\mathbf{x}_{Cu} + \mathbf{x}_{Cl}$, then $x_{Cl} = 0.25$ and $y_{Cl} = 0.14$. Peak II should now check out as the remaining double-weight Cu–Cl interaction at $\mathbf{x}_{Cu} - \mathbf{x}_{Cl}$. Indeed, $\mathbf{x}_{Cu} - \mathbf{x}_{Cl} = \langle -0.17, 0.06 \rangle = -\langle 0.17, -0.06 \rangle$ which agrees tolerably well with the position of peak II. The chlorine position also predicts the position of a peak at $2\mathbf{x}_{Cl}$ with weight 289; peak IV confirms the chlorine assignment. In fact, this Patterson can be solved also for the lighter nitrogen- and carbon-atom positions which account for the remainder of the vectors listed in Table 2.3.2.1. However, the simplest way to complete the structure determination is probably to compute a Fourier synthesis using phases calculated from the heavier copper and chlorine positions.

Consider now a real cell with M crystallographic asymmetric units, each of which contains N atoms. Let us define $\mathbf{x}_{mn}$, the position of the nth atom in the mth crystallographic unit, by

$$\mathbf{x}_{mn} = [\mathbf{T}_m]\mathbf{x}_{1n} + \mathbf{t}_m,$$

where $[\mathbf{T}_m]$ and $\mathbf{t}_m$ are the rotation matrix and translation vector, respectively, for the mth crystallographic symmetry operator. The Patterson of this crystal will contain vector peaks which arise from atoms interacting with other atoms both in the same and in different crystallographic asymmetric units. The set of $(MN)^2$ Patterson vector interactions for this crystal is represented in a matrix in Table 2.3.2.2. Upon dissection of this diagram we see that there are MN origin vectors, $M[(N-1)N]$ vectors from atom interactions with other atoms in the same crystallographic asymmetric unit and $[M(M-1)]N^2$ vectors involving atoms in separate asymmetric units. Often a number of vectors of special significance relating symmetry-equivalent atoms emerge from this milieu of Patterson vectors and such 'Harker vectors' constitute the subject of the next section.

2.3.2.2. Harker sections

Soon after Patterson introduced the F^2 series, Harker (1936) recognized that many types of crystallographic symmetry result in a concentration of vectors at characteristic locations in the Patterson. Specifically, he showed that atoms related by rotation axes produce vectors in characteristic planes of the Patterson, and that atoms related by mirror planes or reflection glide planes produce vectors on characteristic lines. Similarly, noncrystallographic symmetry operators produce analogous concentrations of vectors. Harker showed how special sections through a three-dimensional function could be computed using one- or two-dimensional summations. With the advent of powerful computers, it is usual to calculate a full three-dimensional Patterson synthesis. Nevertheless, 'Harker' planes or lines are often the starting point for a structure determination. It should, however, be noted that non-Harker vectors (those not due to interactions between symmetry-related atoms) can appear by coincidence in a Harker section. Table 2.3.2.3 shows the position in a Patterson of Harker planes and lines produced by all types of crystallographic symmetry operators.

Buerger (1946) noted that Harker sections can be helpful in space-group determination. Concentrations of vectors in appropriate regions of the Patterson should be diagnostic for the presence of some symmetry elements. This is particularly useful where these elements (such as mirror planes) are not directly detected by systematic absences.

Buerger also developed a systematic method of interpreting Harker peaks which he called implication theory [Buerger (1959, Chapter 7)].

Table 2.3.2.2. *Square matrix representation of vector interactions in a Patterson of a crystal with M crystallographic asymmetric units each containing N atoms*

Peak positions $\mathbf{u}_{m1n1, m2n2}$ correspond to vectors between the atoms $\mathbf{x}_{m1n1}$ and $\mathbf{x}_{m2n2}$ where $\mathbf{x}_{mn}$ is the nth atom in the mth crystallographic asymmetric unit. The corresponding weights are $w_{n1}w_{n2}$. The outlined blocks I1 and IM represent vector interactions between atoms in the same crystallographic asymmetric units (there are M such blocks). The off-diagonal blocks IIM1 and II$1M$ represent vector interactions between atoms in crystal asymmetric units 1 and M; there are $M(M-1)$ blocks of this type. The significance of diagonal elements of block IIM1 is that they represent Harker-type interactions between symmetry-equivalent atoms (see Section 2.3.2.2).

	$\mathbf{x}_{11}, w_1$	$\mathbf{x}_{12}, w_2$	$\cdots$	$\mathbf{x}_{1N}, w_N$	$\cdots$	$\mathbf{x}_{M1}, w_1$	$\mathbf{x}_{M2}, w_2$	$\cdots$	$\mathbf{x}_{MN}, w_N$
$\mathbf{x}_{11}, w_1$	$0, w_1^2$	$\mathbf{u}_{11,12}, w_1 w_2$	$\cdots$	$\mathbf{u}_{11,1N}, w_1 w_N$					
$\mathbf{x}_{12}, w_2$		$0, w_2^2$	$\cdots$	$\mathbf{u}_{12,1N}, w_2 w_N$	$\cdots$				
$\vdots$			$\ddots$	$\vdots$					
$\mathbf{x}_{1N}, w_N$				$0, w_N^2$					
	Block I1					Block II$1M$			
$\vdots$		$\vdots$			$\ddots$		$\vdots$		
$\mathbf{x}_{M1}, w_1$	$\mathbf{u}_{M1,11}, w_1^2$	$\mathbf{u}_{M1,12}, w_1 w_2$							
$\mathbf{x}_{M2}, w_2$	$\mathbf{u}_{M2,11}, w_2 w_1$	$\mathbf{u}_{M2,12}, w_2^2$							
$\vdots$			$\ddots$		$\cdots$				
$\mathbf{x}_{MN}, w_N$				$\mathbf{u}_{MN,1N}, w_N^2$					
	Block IIM1					Block IM			

Table 2.3.2.3. *Position of Harker sections within a Patterson*

Symmetry element	Form of $P(x, y, z)$
(*a*) Harker planes	
Axes parallel to the *b* axis:	
(i) 2	$P(x, 0, z)$
(ii) 2_1	$P(x, \frac{1}{2}, z)$
Axes parallel to the *c* axis:	
(i) 2, 3, $\bar{3}$, 4, $\bar{4}$, 6, $\bar{6}$	$P(x, y, 0)$
(ii) 2_1, 4_2, 6_3	$P(x, y, \frac{1}{2})$
(iii) 3_1, 3_2, 6_2, 6_4	$P(x, y, \frac{1}{3})$
(iv) 4_1, 4_3	$P(x, y, \frac{1}{4})$
(v) 6_1, 6_5	$P(x, y, \frac{1}{6})$
(*b*) Harker lines	
Planes perpendicular to the *b* axis:	
(i) Reflection planes	$P(0, y, 0)$
(ii) Glide plane, glide $= \frac{1}{2}a$	$P(\frac{1}{2}, y, 0)$
(iii) Glide plane, glide $= \frac{1}{2}c$	$P(0, y, \frac{1}{2})$
(iv) Glide plane, glide $= \frac{1}{2}(a + c)$	$P(\frac{1}{2}, y, \frac{1}{2})$
(v) Glide plane, glide $= \frac{1}{4}(a + c)$	$P(\frac{1}{4}, y, \frac{1}{4})$
(vi) Glide plane, glide $= \frac{1}{4}(3a + c)$	$P(\frac{3}{4}, y, \frac{1}{4})$
(*c*) Special Harker planes	

Axes parallel to or containing body diagonal (111), valid for cubic space groups only:

            Equation of plane
            $lx + my + nz - p = 0$

(i)   3 $l = m = n = \cos 54.73561° = 0.57735$
                $p = 0$

(ii) 3_1 $l = m = n = \cos 54.73561° = 0.57735$
                $p = \sqrt{3}/3$

Rhombohedral threefold axes produce analogous Harker planes whose description will depend on the interaxial angle.

2.3.2.3. Finding heavy atoms

The previous two sections have developed some of the useful mechanics for interpreting Pattersons. In this section, we will consider finding heavy-atom positions, in the presence of numerous light atoms, from Patterson maps. The feasibility of structure solution by the heavy-atom method depends on a number of factors which include the relative size of the heavy atom and the extent and quality of the data. A useful rule of thumb is that the ratio

$$r = \frac{\sum_{\text{heavy}} Z^2}{\sum_{\text{light}} Z^2}$$

should be near unity if the heavy atom is to provide useful starting phase information (Z is the atomic number of an atom). The condition that $r > 1$ normally guarantees interpretability of the Patterson function in terms of the heavy-atom positions. This 'rule', arising from the work of Luzzati (1953), Woolfson (1956), Sim (1961) and others, is not inviolable; many ambitious determinations have been accomplished *via* the heavy-atom method for which r was well below 1.0. An outstanding example is vitamin B_{12} with formula $C_{62}H_{88}CoO_{14}P$, which gave an $r = 0.14$ for the cobalt atom alone (Hodgkin *et al.*, 1957). One factor contributing to the success of such a determination is that the relative scattering power of Co is enhanced for higher scattering angles. Thus, the ratio, r, provides a conservative estimate. If the value of r is well above 1.0, the initial easier interpretation of the Patterson will come at the expense of poorly defined parameters of the lighter atoms.

A general strategy for determining heavy atoms from the Patterson usually involves the following steps.

(1) List the number and type of atoms in the cell.

(2) Construct the interaction matrix for the heaviest atoms to predict the positions and weights of the largest Patterson vectors. Group recurrent vectors and notice vectors with special properties, such as Harker vectors.

(3) Compute the Patterson using any desired modifications. Placing the map on an absolute scale [$P(000) = \sum Z^2$] is convenient but not necessary.

(4) Examine Harker sections and derive trial atom coordinates from vector positions.

(5) Check the trial coordinates using other vectors in the predicted set. Correlate enantiomorphic choice and origin choice for independent sites.

(6) Include the next-heaviest atoms in the interpretation if possible. In particular, use the cross-vectors with the heaviest atoms.

(7) Use the best heavy-atom model to initiate phasing.

Detailed and instructive examples of using Pattersons to find heavy-atom positions are found in almost every textbook on crystal structure analysis [see, for example, Buerger (1959), Lipson & Cochran (1966) and Stout & Jensen (1968)].

The determination of the crystal structure of cholesteryl iodide by Carlisle & Crowfoot (1945) provides an example of using the Patterson function to locate heavy atoms. There were two molecules, each of formula $C_{27}H_{45}I$, in the $P2_1$ unit cell. The ratio $r = 2.8$ is clearly well over the optimal value of unity. The $P(x, z)$ Patterson projection showed one dominant peak at $\langle 0.434, 0.084 \rangle$ in the asymmetric unit. The equivalent positions for $P2_1$ require that an iodine atom at $x_{\text{I}}, y_{\text{I}}, z_{\text{I}}$ generates another at $-x_{\text{I}}, \frac{1}{2} + y_{\text{I}}, z_{\text{I}}$ and thus produces a Patterson peak at $\langle 2x_{\text{I}}, \frac{1}{2}, 2z_{\text{I}} \rangle$. The iodine position was therefore determined as 0.217, 0.042. The y coordinate of the iodine is arbitrary for $P2_1$ yet the value of $y_{\text{I}} = 0.25$ is convenient, since an inversion centre in the two-atom iodine structure is then exactly at the origin, making all calculated phases 0 or π. Although the presence of this extra symmetry caused some initial difficulties in the interpretation of the steroid backbone, Carlisle and Crowfoot successfully separated the enantiomorphic images. Owing to the presence of the perhaps *too heavy* iodine atom, however, the structure of the carbon skeleton could not be defined very precisely. Nevertheless, all critical stereochemical details were adequately illuminated by this determination. In the cholesteryl iodide example, a number of different yet equivalent origins could have been selected. Alternative origin choices include all combinations of $x \pm \frac{1}{2}$ and $z \pm \frac{1}{2}$.

A further example of using the Patterson to find heavy atoms will be provided in Section 2.3.5.2 on solving for heavy atoms in the presence of noncrystallographic symmetry.

2.3.2.4. Superposition methods. Image detection

As early as 1939, Wrinch (1939) showed that it was possible, in principle, to recover a fundamental set of points from the vector map of that set. Unlike the Harker–Buerger implication theory (Buerger, 1946), the method that Wrinch suggested was capable of using all the vectors in a three-dimensional set, not those restricted to special lines or sections. Wrinch's ideas were developed for vector sets of points, however, and could not be directly applied to real, heavily overlapped Pattersons of a complex structure. These ideas seem to have lain dormant until the early 1950s when a number of independent investigators developed superposition methods (Beevers & Robertson, 1950; Clastre & Gay, 1950; Garrido, 1950*a*; Buerger, 1950*a*).

A Patterson can be considered as a sum of images of a molecule as seen, in turn, for each atom placed on the origin (Fig. 2.3.2.3). Thus, the deconvolution of a Patterson could proceed by superimposing each image of the molecule obtained onto the others by translating the Patterson origin to each imaging atom.
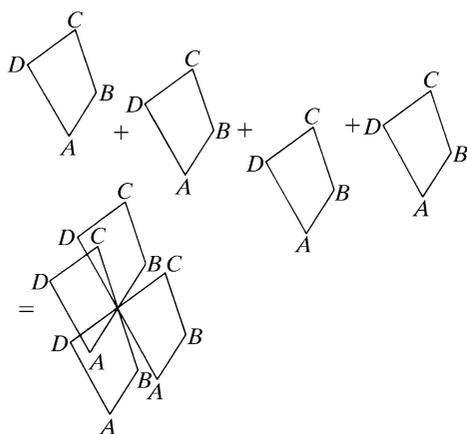
Fig. 2.3.2.3. Atoms *ABCD*, arranged as a quadrilateral, generate a Patterson which is the sum of the images of the quadrilateral when each atom is placed on the origin in turn.

For instance, let us take a molecule consisting of four atoms *ABCD* arranged in the form of a quadrilateral (Fig. 2.3.2.3). Then the Patterson consists of the images of four identical quadrilaterals with atoms *A*, *B*, *C* and *D* placed on the origin in turn. The Pattersons can then be deconvoluted by superimposing two of these Pattersons after translating these (without rotation) by, for instance, the vector **AB**. A further improvement could be obtained by superimposing a third Patterson translated by **AC**. This would have the additional advantage in that *ABC* is a noncentrosymmetric arrangement and, therefore, selects the enantiomorph corresponding to the hand of the atomic arrangement *ABC* [*cf.* Buerger (1951, 1959)].

A basic problem is that knowledge of the vectors **AB** and **AC** also implies some knowledge of the structure at a time when the structure is not yet known. In practice 'good-looking' peaks, estimated to be single peaks by assessing the absolute scale of the structure amplitudes with Wilson statistics, can be assumed to be the result of single interatomic vectors within a molecule. Superposition can then proceed and the result can be inspected for reasonable chemical sense. As many apparently single peaks can be tried systematically using a computer, this technique is useful for selecting and testing a series of reasonable Patterson interpretations (Jacobson *et al.*, 1961).

Three major methods have been used for the detection of molecular images of superimposed Pattersons. These are the sum, product and minimum 'image seeking' functions (Raman & Lipscomb, 1961). The concept of the sum function is to add the images where they superimpose, whereas elsewhere the summed Pattersons will have a lower value owing to lack of image superposition. Therefore, the sum function determines the average Patterson density for superimposed images, and is represented analytically as

$$S(\mathbf{x}) = \sum_{i=1}^{N} P(\mathbf{x} + \mathbf{u}_i),$$

where $S(\mathbf{x})$ is the sum function at $\mathbf{x}$ given by the superposition of the *i*th Patterson translated by $\mathbf{u}_i$, or

$$S(\mathbf{x}) = \sum_{\mathbf{h}} \left\{ F_{\mathbf{h}}^2 \exp(2\pi i \mathbf{h} \cdot \mathbf{x}) \left[\sum_{i=1}^{N} \exp(2\pi i \mathbf{h} \cdot \mathbf{u}_i) \right] \right\}.$$

Setting

$$m \exp(i\varphi_{\mathbf{h}}) = \sum_{i=1}^{N} \exp(2\pi i \mathbf{h} \cdot \mathbf{u}_i)$$

(*m* and $\varphi_{\mathbf{h}}$ can be calculated from the translational vectors used for the superposition),

$$S(\mathbf{x}) = \sum_{\mathbf{h}} F_{\mathbf{h}}^2 m \exp(2\pi i \mathbf{h} \cdot \mathbf{x} + \varphi_{\mathbf{h}}).$$

Thus, the sum function is equivalent to a weighted 'heavy atom' method based on the known atoms assumed by the superposition translation vectors.

The product function is somewhat more vigorous in that the images are enhanced by the product. If an image is superimposed on no image, then the product should be zero.

The product function can be expressed as

$$Pr(\mathbf{x}) = \prod_{i=1}^{N} P(\mathbf{x} + \mathbf{u}_i).$$

When $N = 2$ (**h** and **p** are sets of Miller indices),

$$Pr(\mathbf{x}) = \sum_{\mathbf{h}} \sum_{\mathbf{p}} F_{\mathbf{h}}^2 F_{\mathbf{p}}^2 \exp[2\pi i (\mathbf{h} + \mathbf{p}) \cdot \mathbf{x}]$$
$$\times \exp[2\pi i (\mathbf{h} \cdot \mathbf{u}_i + \mathbf{p} \cdot \mathbf{u}_i)].$$

Successive superpositions using the product functions will quickly be dominated by a few terms with very large coefficients.

Finally, the minimum function is a clever invention of Buerger (Buerger, 1950*b*, 1951, 1953*a*,*b*,*c*; Taylor, 1953; Rogers, 1951). If a superposition is correct then each Patterson must represent an image of the structure. Whenever there are two or more images that intersect in the Patterson, the Patterson density will be greater than a single image. When two different images are superimposed, it is a reasonable hope that at least one of these is a single image. Thus by taking the value of that Patterson which is the minimum, it should be possible to select a single image and eliminate noise from interfering images as far as possible. Although the minimum function is perhaps the most powerful algorithm for image selection of well sharpened Pattersons, it is not readily amenable to Fourier representation.

The minimum function was conceived on the basis of selecting positive images on a near-zero background. If it were desired to select negative images [*e.g.* the $(\mathbf{F}_1 - \mathbf{F}_2)^2$ correlation function discussed in Section 2.3.3.4], then it would be necessary to use a maximum function. In fact, normally, an image has finite volume with varying density. Thus, some modification of the minimum function is necessary in those cases where the image is large compared to the volume of the unit cell, as in low-resolution protein structures (Rossmann, 1961*b*). Nordman (1966) used the average of the Patterson values of the lowest 10 to 20 per cent of the vectors in comparing Pattersons with hypothetical point Pattersons. A similar criterion was used by High & Kraut (1966).

Image-seeking methods using Patterson superposition have been used extensively (Beevers & Robertson, 1950; Garrido, 1950*b*; Robertson, 1951). For a review the reader is referred to *Vector Space* (Buerger, 1959) and a paper by Fridrichsons & Mathieson (1962). However, with the advent of computerized direct methods (see Chapter 2.2), such techniques are no longer popular. Nevertheless, they provide the conceptual framework for the rotation and translation functions (see Sections 2.3.6 and 2.3.7).
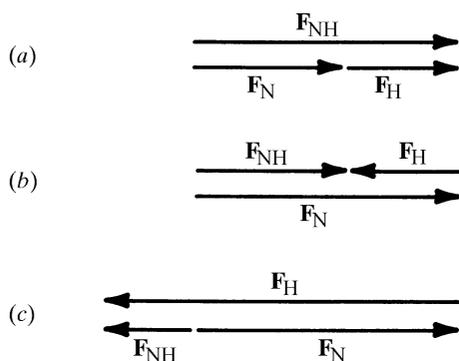
Fig. 2.3.3.1. Three different cases which can occur in the relation of the native, $\mathbf{F}_N$, and heavy-atom derivative, $\mathbf{F}_{NH}$, structure factors for centrosymmetric reflections. $\mathbf{F}_N$ is assumed to have a phase of 0, although analogous diagrams could be drawn when $\mathbf{F}_N$ has a phase of π. The crossover situation in (c) is clearly rare if the heavy-atom substitution is small compared to the native molecule, and can in general be neglected.

2.3.2.5. Systematic computerized Patterson vector-search procedures. Looking for rigid bodies

The power of the modern digital computer has enabled rapid access to the large number of Patterson density values which can serve as a lookup table for systematic vector-search procedures. In the late 1950s, investigators began to use systematic searches for the placement of single atoms, of known chemical groups or fragments and of entire known structures. Methods for locating single atoms were developed and called variously: vector verification (Mighell & Jacobson, 1963), symmetry minimum function (Kraut, 1961; Simpson et al., 1965; Corfield & Rosenstein, 1966) and consistency functions (Hamilton, 1965). Patterson superposition techniques using stored function values were often used to image the structure from the known portion. In such single-site search procedures, single atoms are placed at all possible positions in a crystal, using a search grid of the same fineness as for the stored Patterson function, preferably about one-third of the resolution of the Patterson map. Solutions are gauged to be acceptable if all expected vectors due to symmetry-related atoms are observed above a specified threshold in the Patterson.

Systematic computerized Patterson search procedures for orienting and positioning known molecular fragments were also developed in the early 1960s. These hierarchical procedures rely on first using the 'self'-vectors which depend only on the orientation of a molecular fragment. A search for the position of the fragment relative to the crystal symmetry elements then uses the cross-vectors between molecules (see Sections 2.3.6 and 2.3.7). Nordman constructed a weighted point representation of the predicted vector set for a fragment (Nordman & Nakatsu, 1963; Nordman, 1966) and successfully solved the structure of a number of complex alkaloids. Huber (1965) used the convolution molecule method of Hoppe (1957a) in three dimensions to solve a number of natural-product structures, including steroids. Various program systems have used Patterson search methods operating in real space to solve complex structures (Braun et al., 1969; Egert, 1983).

Others have used reciprocal-space procedures for locating known fragments. Tollin & Cochran (1964) developed a procedure for determining the orientation of planar groups by searching for origin-containing planes of high density in the Patterson. General procedures using reciprocal-space representations for determining rotation and translation parameters have been developed and will be described in Sections 2.3.6 and 2.3.7, respectively.

Although as many functions have been used to detect solutions in these Patterson search procedures as there are programs, most rely on some variation of the sum, product and minimum functions (Section 2.3.2.4). The quality of the stored Patterson density representation also varies widely, but it is now common to use 16

or more bits for single density values. Treatment of vector overlap is handled differently by different investigators and the choice will depend on the degree of overlapping (Nordman & Schilling, 1970; Nordman, 1972). General Gaussian multiplicity corrections can be employed to treat coincidental overlap of independent vectors in general positions and overlap which occurs for symmetric peaks in the vicinity of a special position or mirror plane in the Patterson (G. Kamer, S. Ramakumar & P. Argos, unpublished results; Rossmann et al., 1972).

2.3.3. Isomorphous replacement difference Pattersons

2.3.3.1. Introduction

One of the initial stages in the application of the isomorphous replacement method is the determination of heavy-atom positions. Indeed, this step of a structure determination can often be the most challenging. Not only may the number of heavy-atom sites be unknown, and have incomplete substitution, but the various isomorphous compounds may also lack isomorphism. To compound these problems, the error in the measurement of the isomorphous difference in structure amplitudes is often comparable to the differences themselves. Clearly, therefore, the ease with which a particular problem can be solved is closely correlated with the quality of the data-measuring procedure.

The isomorphous replacement method was used incidentally by Bragg in the solution of NaCl and KCl. It was later formalized by J. M. Robertson in the analysis of phthalocyanine where the coordination centre could be Pt, Ni and other metals (Robertson, 1935, 1936; Robertson & Woodward, 1937). In this and similar cases, there was no difficulty in finding the heavy-atom positions. Not only were the heavy atoms frequently in special positions, but they dominated the total scattering effect. It was not until Perutz and his colleagues (Green et al., 1954; Bragg & Perutz, 1954) applied the technique to the solution of haemoglobin, a protein of 68 000 Da, that it was necessary to consider methods for detecting heavy atoms. The effect of a single heavy atom, even uranium, can only have a very marginal effect on the structure amplitudes of a crystalline macromolecule. Hence, techniques had to be developed which were dependent on the difference of the isomorphous structure amplitudes rather than on the solution of the Patterson of the heavy-atom-derivative compound on its own.

2.3.3.2. Finding heavy atoms with centrosymmetric projections

Phases in a centrosymmetric projection will be 0 or π if the origin is chosen at the centre of symmetry. Hence, the native structure factor, $\mathbf{F}_N$, and the heavy-atom-derivative structure factor, $\mathbf{F}_{NH}$, will be collinear. It follows that the structure amplitude, $|\mathbf{F}_H|$, of the heavy atoms alone in the cell will be given by

$$|\mathbf{F}_H| = |(|\mathbf{F}_{NH}| \pm |\mathbf{F}_N|)| + \varepsilon,$$

where ε is the error on the parenthetic sum or difference. Three different cases may arise (Fig. 2.3.3.1). Since the situation shown in Fig. 2.3.3.1(c) is rare, in general

$$|\mathbf{F}_H|^2 \simeq (|\mathbf{F}_{NH}| - |\mathbf{F}_N|)^2. \tag{2.3.3.1}$$

Thus, a Patterson computed with the square of the differences between the native and derivative structure amplitudes of a centrosymmetric projection will approximate to a Patterson of the heavy atoms alone.

The approximation (2.3.3.1) is valid if the heavy-atom substitution is small enough to make $|\mathbf{F}_H| \ll |\mathbf{F}_{NH}|$ for most reflections, but sufficiently large to make $\varepsilon \ll (|\mathbf{F}_{NH}| - |\mathbf{F}_N|)^2$. It is also

references