A Metropolis-Like Algorithm to Improve Phases: Some Preliminary Results

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Abstract

A procedure to improve phases by a Metropolis-like algorithm (MLA) in reciprocal space is presented. It uses a density modification method (DMM) in a MLA to evaluate progress and to suggest changes to phase angles. First, in the neighborhood of the trial values derived from a DMM, new options for the phase angles are generated. Then, a DMM is used to evaluate and improve compliance with the rule of non-negativity of direct methods. The procedure of DMM → phase change → DMM → phase change is iterated several times. For a centrosymmetric case in which phases may not be changed by a DMM, compliance with the non-negativity rule is judged by the ratio of observed to calculated structure-factor amplitudes, and new options for the phase angles are generated by adding 180°. For a non-centrosymmetric case both the amplitude and the phase values of the structure factors before and after a DMM may differ. However, only the phase values are used to judge the compliance with the non-negativity rule, and to decide phase shifts in a subsequent phase change step. This procedure is shown to improve the phases determined *ab initio* by the consistent electron density approach [Bhat (1984). *Acta Cryst.* A**40**, C15; (1985). Am. Crystallogr. Assoc. Annu. Meet., Stanford, California. Abstr. H1]. The mean phase errors for the phases obtained by the procedure proposed here ranged from 55 to 25° in different tests, significantly better than those obtained from a DMM under similar conditions.

Introduction

The Metropolis algorithm (Metropolis, Rosenbluth, Rosenbluth, Teller & Teller, 1953) (also known as simulated annealing or dynamics with 'slow cooling') may be thought of as an algorithm for very large-scale problems of non-linear equations (Press, Flannery, Teukolsky & Vetterling, 1988). Simulated annealing is specially designed for cases which lead to several local minima. Other methods, such as ones using derivatives, such as Newton's, or conjugate gradients may be compared to 'rapid cooling'. These methods look for nearby minima, which may be found quickly but may not be the lowest (Press *et al.*, 1988). By contrast, simulated annealing takes 'downhill' steps while sometimes allowing 'uphill' steps so as to skip

a barrier giving rise to a local minimum. In this algorithm, shifts are not obtained from the derivatives and hence the corrections need not stop when the derivatives tend to zero at a local minimum. Here its use is proposed to improve tentative phase angles in a phase improvement procedure (PIP). The use of the PIP to improve *ab initio* phases determined by the consistent electron density approach (CEDA; Bhat, 1984, 1985) is illustrated below for both centrosymmetric and non-centrosymmetric cases. These results from PIP are compared with those obtained from the use of a DMM alone.

The method

The basic steps of the Metropolis-like algorithm (MLA) proposed here in a phase improvement procedure are:

(1) description of a possible configuration of the system;

(2) generation of changes in that configuration; these changes are the options presented to the system;

(3) correction of non-beneficial changes made in step (2); and

(4) use of an appropriate annealing schedule, with a control parameter T (analog of 'temperature') for step (2) above.

In the PIP, progress is evaluated using a DMM (Hope & Gassman, 1968) in step 3. If this DMM is adequate to distinguish beneficial from non-beneficial changes, the result of step 2 above, followed by the DMM, will be an improved result.

Different generators of phase changes are used in step 2 for the two symmetry cases. For a centrosymmetric case the phases are restricted to one of the two possible values, and a DMM may not be able to effect such large phase changes. Hence, the compliance with a density modification function in step 3 cannot be judged from the phase change due to a DMM. Therefore, the compliance with the density modification function is judged by the ratio of the structure-factor amplitudes before and after a DMM. Thus in step 2 above, following a DMM phase angles are changed by 180° for reflections showing a poor ratio of calculated to observed structure-factor amplitudes. The success of such a change is again evaluated using a DMM, and the process of phase change and its evaluation is iterated several times (see below).

For a non-centrosymmetric case both the ratio of the structure-factor amplitudes and the phase changes due to a DMM may be used to estimate the compliance with the density modification function. However, because of lack of restriction (*e.g.* 0 or 180°) on phases, the ratio of the amplitudes may be used in the phase-change step 2 only through a function relating that ratio to a possible phase error. Therefore, in this preliminary effort to develop the PIP, only the phase changes due to a DMM are used to estimate the compliance, and thus to decide shifts in step 2 of the algorithm.

The true minimum is distinguished from a false minimum by its behavior. The true minimum will be more pronounced and sustained throughout than a false minimum. Thus, we assume that, near the right minimum, the net phase change from a phase change step and a DMM is negligible; that is, a cancelling-out effect ('complementarity') will be seen between the phase shifts from these two steps. A lack of such 'complementarity' in phase shifts from these two steps is taken as an indication that the phase is not near the true minimum. Therefore, phases showing a poorer 'complementarity' are given larger changes in step 2.

For a non-centrosymmetric case the 'complementarity' notion and for the centrosymmetric case the ratios of calculated to observed structure factors are used to introduce phase changes. It is conceivable that one might combine these two approaches. For example, estimates of phase errors from observed and calculated structure-factor amplitudes (Sim, 1959) can be used with or without the 'complementarity' notion to make changes in step 2.

The true minimum may be aimed at in a PIP only if (*a*) the DMM accurately expresses the properties of the true electron density distribution, and (*b*) if the shifts in the phase-change step 2 are small enough to be contained by the true minimum, but large enough to overcome all the local minima. In practical cases, the above condition is hard to satisfy. Thus one may be looking for a minimum which cannot be significantly improved with the available information, and the PIP may only aim at a workable rather than the global minimum.

The steps of the procedure

The PIP is an iterative procedure involving a step to identify new options for the phase values [(*d*) below] within or beyond tentative minima, and then [steps (*a*) to (*c*) below] to estimate and improve the compliance with the expected property of the electron density using DMM. The sequence of steps is as follows:

(*a*) from initial phases φ and the observed amplitudes $|F_o|$ an electron density map ρ is calculated;

(*b*) the map, ρ, is modified using a DMM;

(*c*) the modified map is inverse-Fourier transformed to obtain structure-factor amplitudes $|F_c|$ and phases φ';

(*d*) phases φ' are altered from their tentative values for selected (centrosymmetric) or for all (non-centrosymmetric) reflections as described later. This step (performing step 2 in the method) forces some 'uphill' shifts in a DMM result, and thus permits the exploration of other options for the phase angles in and around the available values. It provides options to verify whether the latest minima for the phases are stable to changes, as true minima should be, or are merely local minima. By systematically testing other values for the phases, it seeks an answer to the question: can we further improve the compliance with the density modification function?;

(*e*) from the altered phases and the $|F_o|$ a new electron density map is calculated as in (*a*) above.

Density modification [step (*b*)]

The physical properties of the electron density are introduced in a DMM (i) using the observed amplitudes, and (ii) through a density modification step. The most fundamental of the features used in a DMM is that of the non-negativity rule (Karle, 1985; Karle & Hauptman, 1950; Sayre, 1952; Woolfson, 1987) employed by direct methods. This is used here as follows:

$$\begin{aligned} \rho' &= \rho & \text{if } \rho \geq 0 \\ \rho' &= \rho * S, & \text{if } \rho < 0, \end{aligned} \quad (1)$$

where S is a positive constant (<1), which is related to the maximum resolution of the data. In the absence of series termination and when $F(000)$ is included, S is zero at the final cycles of the PIP. For smooth convergence, the MLA requires that the shifts be small enough to remain in a global minimum. Since the shifts are dependent on the value of S, S is chosen to give a small R factor (*e.g.* around 10%) between the structure-factor amplitudes calculated from ρ and ρ'.

To comply with the non-negativity rule of the electron density, the following modifications are often used in a DMM:

$$\begin{aligned} \rho' &= \rho, & \text{if } \rho \geq C \\ \rho' &= C, & \text{if } \rho < 0, \end{aligned} \quad (2)$$

where C is a small constant for small molecules or the solvent level in crystals of macromolecules.

In macromolecular crystals at low or medium resolution, not only the errors in the phase angles, but also series termination may lead to negative electron density values. If the $F(000)$ term is omitted, electron density values within a molecular boundary show greater positive and negative excursions than in the relatively featureless solvent regions (Reynolds *et al.*,

1985). A DMM which uses (2) resets large negative values to a constant. Such an approach ignores the presence of larger negative values due to series termination errors. The modification (1) lacks this limitation as it only tries to attenuate large negative values. Thus, this approach permits the co-appearance of negative values arising from series termination along with zero or smaller negative values due to solvent and other factors.

Phase change [step (d)]

Because of the restriction on phase angles by symmetry, this step differs for centrosymmetric and non-centrosymmetric space groups. For centrosymmetric cases, the phases φ' are modified in step (d) such that

$$\begin{aligned} \varphi'' &= \varphi' \pm 180° \quad \text{if } |F_c|/|F_o| < T \\ \varphi'' &= \varphi' \qquad\quad \text{if } |F_c|/|F_o| \geq T, \end{aligned} \quad (3)$$

where T is a threshold value chosen such that a small initial number, e.g. 15%, of the total reflections have $|F_c|/|F_o| < T$. As the iteration proceeds, the number of phases affected by (3) is reduced by changing T. This step may also be used in a non-centrosymmetric space group for centric reflections that have special phase values (e.g. $\pi/2, 3\pi/2$ in $P2_12_12_1$). The use of $|F_o| - |F_c|$ instead of $|F_c|/|F_o|$ will not only make T sensitive to a scale factor between $|F_o|$ and $|F_c|$ but will also make the change by (3) to be dominated by the stronger low-resolution amplitudes. Use of the threshold ratio for the inverse of $|F_c|/|F_o|$ in (3) is not consistent with the statistical derivations by Woolfson (1956) and by Sim (1959). Replacement of the threshold value by a probability function using the expression derived by Woolfson (1956) is a possible modification to (3).

For non-centrosymmetric space groups φ'' is obtained as

$$\varphi'' = \varphi' + RD\delta\varphi + \Delta\varphi. \quad (4)$$

D is a damping factor chosen such that the mean difference between φ'' and φ' is small, e.g. 10°. An excessively large value for D would lead to poor convergence of the phases, whereas too small a value for D might fail to take the phases out of a local minimum. $\delta\varphi$ is the lack of 'complementarity' from the previous cycle. $\delta\varphi = 0$ corresponds to perfect 'complementarity', that is, the phase angle sustained the perturbations due to the phase change step in the previous cycle. R is a random number to explore different possibilities during a MLA to improve compliance with the density modification function. This may be compared with the random number used in molecular-dynamics simulations to propagate random motion prior to an optimization cycle involving stereochemical restraints. In this sense, the phase-change step (d) can be called a 'dynamics' step in

which the phase 'move' in phase space constrained by a density modification function. $\Delta\varphi$ is a constant chosen such that φ'' and φ' differ from each other even when $\delta\varphi$ is zero. $\Delta\varphi$ may also be used to increase 'motion' when phases have converged to a local minimum.

Comparison of centro- and non-centrosymmetric cases

Equation (3) for phase change in a centrosymmetric case is a special case of (4) for the non-centrosymmetric case as shown below. For smooth convergence, the DMM may introduce only small changes in each cycle of a PIP. Because the centric phases calculated from two very similar densities are likely to be the same, in a PIP the centrosymmetric phases may not change through a DMM. Thus for a centrosymmetric case, (4) reduces ($\delta\varphi = 0$) to (3). Because $\Delta\varphi$ can only be large (180°) for a centrosymmetric case, in order to maintain a small mean phase change in (3) modification is done only for a selected number of reflections. A Metropolis algorithm would require this selection to be made such that the number of reflections changing phase in (3) form a Boltzmann distribution with respect to an 'energy value' associated with $|F_c|/|F_o|$. This has not been verified in the method proposed here, which for this reason is called a Metropolis-like, rather than Metropolis, algorithm.

The phase improvement procedure (PIP) versus Monte Carlo and molecular-dynamics simulations

The implementation of a MLA in the PIP is close in spirit to the Monte Carlo and molecular-dynamics simulations. In Monte Carlo, the acceptance of shifts (generated randomly) is determined by a Boltzmann distribution factor, whereas in molecular-dynamics simulations, random shifts are propagated by solving Newton's equations of motion. In PIP, the shifts are simulated so as to satisfy the expected density modification functions. Thus PIP uses a MLA with the expected density modification functions to survey possible options for the phases. The molecular-dynamics and Monte Carlo simulations use the Metropolis algorithm with stereochemical restrictions to examine possible spatial rearrangements. By analogy, the effect of different 'temperatures', if required, may be introduced by suitably changing the density modification functions. Small modifications [that is $S \sim 1$ in (1)] in a DMM will lead to smaller shifts in (4).

Molecular dynamics is a useful technique for the crystallographic refinement of macromolecules (Brünger, Kuriyan & Karplus, 1987; van Gunsteren & Berendsen, 1987; Fujinaga, Gros & van Gunsteren, 1989). It may be used only when an atomic model is available. The PIP is model independent, that is, the shifts are directly applied to the phases. However, it

must be noted that the results discussed in this article are based on calculations with crystals of small molecules. The usefulness of the PIP for crystals of macromolecules has not been demonstrated.

The energy barriers for molecular motions arise from factors such as non-bonded contacts and charge–charge interactions. Molecular dynamics overcomes these barriers by introducing motion away from the trial positions. The PIP similarly attempts to overcome causes preventing change in phase angles during DMM and OMIT map [an electron density map calculated from the observed amplitudes using phases from all but the atoms within and around a particular volume (Artymiuk & Blake, 1981; Bhat & Cohen, 1984; Bhat, 1988)] cycles of the consistent electron density approach (CEDA; Bhat, 1984, 1985). Phase angles of low resolution or centric reflections may not be readily changed by a DMM or by an OMIT map as these reflections may not sense small local changes in the electron density values. There-fore, a MLA or 'dynamics' is proposed here to make it easier to overcome these limitations of DMM and OMIT map procedures.

Implementation of the Metropolis-like algorithm in the phase improvement procedure (PIP)

Equations (3) and (4) used for the two symmetry cases can be compared to the elements of the Metropolis algorithm as follows. The use of the Metropolis algorithm, for example, in the travelling-salesman problem (Press et al., 1988) assumes that the system can be grouped into several smaller sec-tions. Each section may be given new options and a penalty function can be calculated. From this func-tion, the method accepts or rejects the suggested option. The Metropolis algorithm also assumes that, for a given section, the number of options is finite. Similarly in the phase problem each reflection may be treated as a section. For centrosymmetric reflec-tions, the options are indeed finite (0 or 180°), and these two possibilities may be tried one by one for reflections with poor $|F_c|$ to $|F_o|$ ratio [(3)] after the inverse Fourier transforms of a modified electron-density map. However, for the non-centrosymmetric case, possible values for phase are infinite, and it is inconceivable to try all of them; the $|F_c|/|F_o|$ ratio does not right away suggest new options for the phase angles either. Therefore, the 'complementarity' notion is introduced to obtain [(4)] the trial values for explor-ing other possibilities of the phase angles in step 2. Our results from (4) are encouraging (see below) when used with reasonable starting phases for a small molecule. However, as discussed later, the chances of reaching a workable minimum depend on the tenta-tive starting phases. Therefore, the validity of such a 'complementarity' notion for different starting condi-tions needs further thought and experimentation.

Table 1. *Phase errors as a function of resolution after* 0, 10, 20, 30 *cycles of the phase improvement procedure at* 1·5 Å *resolution* $(E > 1)$ *for HPATT*

Starting phases were obtained by the CEDA (Bhat *et al.*, 1990).

Cycle number	Phase error (°) from ∞ to resolution (Å)					
	5·0	3·0	2·5	2·0	1·7	1·5
0	0	54	49	65	88	83
10	0	72	65	56	73	77
20	0	36	33	35	62	69
30	0	18	16	29	48	52
Number of reflections	2	10	11	25	49	65

Application for a centrosymmetric case

The structure of 5-hydroxy-5-phenyl-7-azatricyclo-[7.4.0.0^{2,7}]trideca-2,9(1),10,12-tetraen-8-one (HPATT; Bhat, Ammon, Mazzocchi & Oda, 1990) was solved from an E map calculated with phases determined *ab initio* by the consistent electron density approach (CEDA). The mean phase difference (phase error) between the phases determined by the CEDA and those from the final refined model was 49° to 2·5 Å, and 83° for data to 1·5 Å resolution. All E values ≥ 1 were used. The space group is $P2_1/n$, with $a = 7·408$, $b = 22·311$, $c = 8·613$ Å, $\beta = 103·53°$. The molecular formula is $C_{18}H_{15}NO_2$, with one molecule per asymmetric unit. The structure was refined to an R factor of 0·05 for the 963 $|F_o|$.

The phase values determined [prior to the model building step of Bhat *et al.* (1990)] by the CEDA for reflections with $E \geq 1$, out to 1·5 Å, were used as the starting phases for a PIP. The routines described by Bhat (1988) were used for the calculation of both the forward and the inverse Fourier transforms. These transforms used 30, 90 and 36 grid divisions along **a**, **b** and **c** respectively. The PIP was applied in the following three stages. [These three stages may be compared to the simulated annealing technique of a Metropolis procedure which initially permits large changes (heating) and then gradually limits them (cooling) as the cycle proceeds].

Stage 1: From the phases determined by the CEDA an E map was calculated for all the reflections out to 1·5 Å with $E \geq 1$. Values in this E map were modified using (1) with $S = 0·35$. The E map was inverse Fourier transformed to get $|E_c|$ giving an R factor of 0·07 with

$$R = \sum \|E_c| - k|E_o\|/\sum |E_o|, \qquad (5)$$

where k is a scale factor. At this stage, for the 1·5 Å data set the error in φ' was 83° (Table 1). About 20% of the total reflections had an $|E_c|/|E_o|$ ratio of less than 0·65. Therefore 0·65 was chosen as the value of T in (3) and ten cycles of the PIP [steps (*a*) to (*d*)] were performed. During these cycles, the phase error for the 1·5 Å data set improved from 83 to 77° (Table 2). During the last few cycles the phase error oscil-lated between 69 and 80°.

Table 2. *Errors in phase angles after the phase improvement procedure for HPATT using the 1·5 Å data set (E > 1)*

Ab initio phases were obtained by the CEDA (Bhat *et al.*, 1990).

Cycle number	Phase error (°) to 1·5 Å
1	83
2	86
3	86
4	75
5	75
6	72
7	72
8	80
9	69
10	77
11	69
12	66
13	66
14	69
15	61
16	69
17	61
18	69
19	61
20	69
21	61
22	64
23	61
24	64
25	55
26	55
27	52
28	55
29	55
30	52

Stage 2: Ten additional PIP cycles were done choosing T such that about ten phases were changed by (3) in each cycle. This reduced the phase error to 69°; it ranged from 61 to 69° in the last few cycles.

Step 3: Ten further cycles were performed choosing T such that only about five phases were changed in step (d) by (3), and the error decreased to about 52°. During the last few cycles the phase error ranged from 52 to 55° (Table 2) for all the reflections out to 1·5 Å resolution with $E > 1$. The corresponding value for all the reflections to 2·5 Å resolution is 16° which is a decrease of 33° from the initial error. The phase angles at the end of 0, 10, 20 and 30 cycles are given in Table 3.

Use of the phase improvement procedure in a non-centrosymmetric case

The structure of N,N,N',N'-tetrakis(2-fluoro-2,2-dinitroethyl)oxamide (TO) has been solved (Bhat & Ammon, 1987, 1990) using phases determined by the CEDA. The phase error was 67° for all the reflections out to 3 Å with observed structure-factor amplitude ≥ 170. The molecular formula is $C_{10}H_8F_4N_{10}O_{18}$, with one molecule per asymmetric unit. The space group is Pc with $a = 7·88$, $b = 6·778$, $c = 21·595$ Å, $\beta = 108·21°$. The structure was refined to an R factor of

Table 3. *Reflection data and phase angles after 0, 10, 20 and 30 cycles of the phase improvement procedure for HPATT using the 1·5 Å data set (E > 1)*

Starting phases were obtained by the CEDA (Bhat *et al.*, 1990).

h	k	l	$|E_o|$	Phases (°) in cycle number 0	10	20	30
-4	0	4	1·19	180	180	180	180
-4	3	1	1·87	0	0	0	0
-4	3	4	1·17	180	0	0	0
-4	4	1	1·25	180	180	0	0
-4	5	1	1·31	180	180	180	180
-4	6	2	1·25	180	180	0	0
-3	1	1	1·13	0	180	180	180
-3	3	3	1·45	180	180	180	180
-2	2	1	1·08	0	0	0	0
-2	3	4	1·36	180	180	180	180
-2	5	4	1·40	180	180	180	180
-2	6	5	1·20	180	180	0	0
-2	7	3	1·12	180	0	0	0
-2	9	2	1·23	0	0	0	0
-2	10	2	1·41	0	0	0	180
-2	12	1	1·05	180	180	180	180
-1	1	4	1·51	180	180	180	180
-1	2	1	1·04	0	0	0	0
-1	2	5	1·21	180	0	180	0
-1	5	3	1·55	180	180	180	180
-1	6	3	1·87	180	180	180	180
-1	6	4	1·20	180	180	180	180
-1	9	3	1·17	0	180	0	180
-1	10	1	1·09	180	0	0	0
-1	12	1	1·51	0	0	0	0
-1	12	3	1·11	0	180	0	180
0	3	1	1·27	0	0	0	0
0	4	2	1·46	180	180	180	180
0	4	4	1·20	0	0	0	0
0	6	1	1·22	180	0	180	180
0	6	4	1·11	180	0	180	180
0	8	3	1·41	180	180	180	180
0	9	1	1·72	180	0	0	0
0	9	2	1·60	180	180	180	180
0	10	0	1·31	180	180	180	180
0	10	3	1·31	0	0	0	0
0	11	1	1·14	180	180	180	180
1	-10	0	1·07	180	180	180	180
1	0	3	1·91	0	0	180	180
1	0	5	1·44	0	0	0	0
1	2	5	1·05	180	180	180	180
1	3	4	1·54	0	0	0	0
1	4	0	1·03	180	180	180	180
1	5	3	1·55	0	0	0	0
1	6	3	1·63	180	0	0	0
1	8	2	1·40	0	0	0	0
1	10	3	1·39	0	180	0	0
1	11	3	1·18	0	180	180	180
2	-5	0	1·02	180	180	180	180
2	-4	0	1·27	0	0	0	180
2	-3	0	1·62	180	180	180	180
2	-2	0	1·97	0	0	180	180
2	0	0	2·39	180	180	180	180
2	4	2	1·32	0	0	0	0
2	6	4	1·19	0	180	0	0
2	9	3	1·67	180	180	0	180
3	3	3	1·07	180	0	180	180
3	5	3	1·01	180	180	180	180
3	7	0	1·28	180	0	180	180
4	-7	0	1·04	180	180	0	0
4	-6	0	1·19	0	0	0	0
4	-4	0	1·61	180	0	180	180
4	-3	0	2·53	180	180	0	0
4	0	0	1·31	180	0	0	0
4	1	0	1·38	0	0	180	180

Table 4. *Errors in phase angles after the phase improvement procedure, and density modification cycles for TO*

Ab initio phases were obtained by the CEDA (Bhat & Ammon, 1990).

Cycle number	Phase error (°) to 3 Å	
	PIP	DMM
0	66·88	66·88
1	66·88	66·46
2	66·90	66·16
3	66·81	65·98
4	66·84	65·87
5	66·76	65·87
6	66·78	65·84
7	66·70	65·57
8	57·15	65·28
9	58·41	64·99
10	56·99	64·75
11	56·15	64·74
12	57·36	64·67
13	55·59	64·32
14	56·93	63·97
15	55·97	63·76
16	55·61	63·75
17	55·47	63·74
18	52·25	63·73
19	55·26	63·46
20	49·26	63·36
21	52·69	63·35
22	46·90	63·33

Table 5. *Initial and final phase errors (°) after 20 cycles of the phase improvement procedure for HPATT*

Initial phases were obtained by introducing random errors to the phases obtained *ab initio* by the CEDA followed by the phase improvement procedure.

Set	Initial error	Final error
1	75	53
2	69	41
3	77	55
4	75	30
5	64	25

and the first five sets which had phase error of <80° were improved (Table 5) in 20 cycles of stage 1. T was chosen so as to change the sign of about two reflections in each cycle. In all these calculations the accuracy of the phases improved significantly; in one case the error was only 25° from the phases of the refined final atomic model (Fig. 1). However, calculations initiated with a mean phase error of about 90° lead to only marginal improvements (about 10°) under similar conditions. These calculations suggest that an initial set of non-random phases (from a CEDA in the test calculations) are necessary for the success of the method when used with the minimization function suggested in this paper. The possibility of choosing functional forms different from (4) has not been studied. A function based on maximum entropy is a possibility. These calculations took about 4 CPU min per cycle on a VAXstation 3500.

0·047 for the 1733 $|F_o|$. The phases determined (prior to the model-building step of Bhat & Ammon, 1990) *ab initio* by the CEDA were improved by PIP as follows.

Initial phases for TO were better than those of HPATT, hence an annealing schedule with varying D and $\Delta\varphi$ was not used for TO. 22 cycles of PIP were performed with $S = 0·9$ in (1) and $D = 0·5$, $\Delta\varphi = 0·02 \times 360°$ in (4). These calculations used 28, 26 and 76 grid divisions along the a, b and c axes respectively. The phase error for the 3 Å data set decreased from 67 to 47° (Table 4) and during the last few cycles it oscillated between 47 and 52°.

False minima and convergence

Success of a MLA depends on the validity of the approaches for the phase change and progress evaluation steps (2 and 3). In our tests with the PIP, shifts (step 2) were dependent on the compliance with the non-negativity rule imposed by a DMM. However, the DMM itself is based on the tentative electron density. Therefore, the density modification functions leading to the final minimum are not 'absolute', that is, they are not independent of the tentative phases. Such a dependence between the functions defining the final minimum and the initial density values itself may cause problems, that is, step (b) may not necessarily distinguish beneficial from non-beneficial changes made by step (d) when the starting phases are far from the true minimum. The following tests were done to study such correlations.

The phases obtained by the method for the centrosymmetric case (HPATT) were randomly altered

The phase improvement procedure *versus* a density modification procedure

For a given reflection, improvement in the ratio of $|F_c|$ to $|F_o|$ is likely to be seen even at a false minimum, and since there are many false minima and only one right one, results of a DMM are likely to be influenced by false minima. However, a local minimum will be less pronounced (smaller ratio of $|F_c|$ to $|F_o|$, poorer 'complementarity') than the global minimum. The PIP exploits this unique property of the true minimum to aim at better results. The following tests were done with HPATT and TO to compare a DMM with the PIP. For these structures, the phases determined by the CEDA were not the best that could be obtained but the calculation was stopped when the map was good enough to be interpreted. Therefore, additional DMM cycles were performed with the same parameters used in PIP. These calculations to improve further the phases determined by the CEDA used a DMM:

$$|F_o|, \varphi \xrightarrow[\text{FFT}]{} \rho \xrightarrow[\substack{\text{density} \\ \text{modification}}]{} \rho' \xrightarrow[\text{FFT}]{} |F_c|, \varphi' \xrightarrow[\text{FFT}]{} \rho. \tag{6}$$

The density modification steps were as outlined in (1).

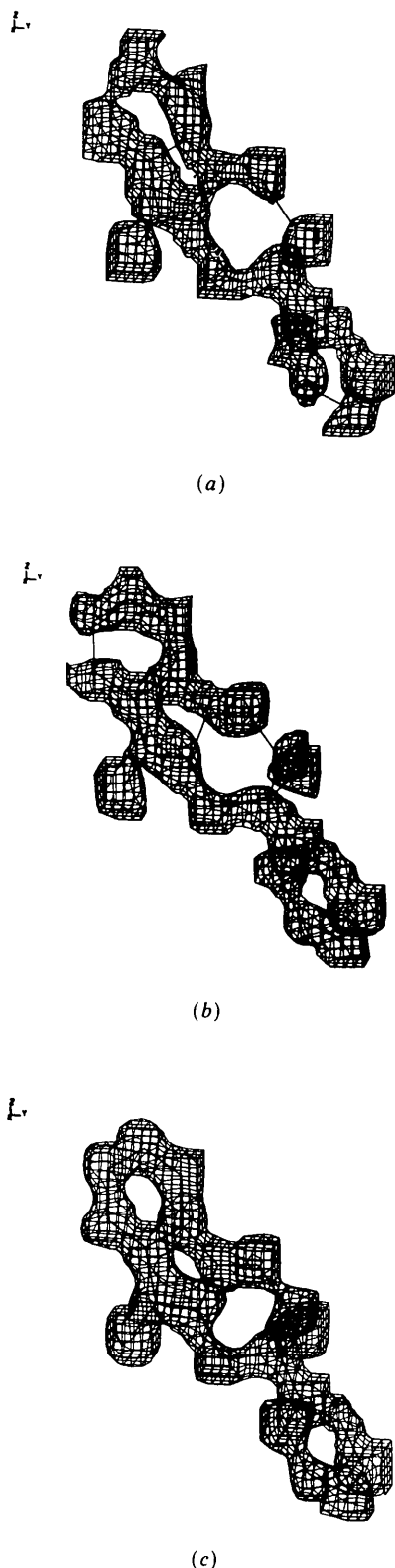The procedure (6) did not improve the phases for the centrosymmetric case (HPATT) described above.

(a)

(b)

(c)

Fig. 1. Electron density maps for HPATT with phases from the phase improvement procedure. (a) Phase error of 42°; (b) phase error of 25°. (c) Phases from the refined model. The atomic model is also shown. The same contour level is used in all the figures.

This lack of progress by the DMM may be explained as follows. Centrosymmetric reflections pose a special problem for phase refinement methods such as DMM or the CEDA. These methods improve the phases by gradually shifting their initial phase angles (Bhat *et al.*, 1990), which is not possible if the phases are restricted to 0 or 180°.

For the non-centrosymmetric case (TO) the phases determined by the CEDA could be improved by the DMM alone (Table 4) from 67 to 63° in 22 cycles. The value of S in (1) and the sampling interval in the Fourier transforms were the same for both the PIP and the DMM. Under these identical conditions, the DMM improved the phases by only 4° whereas the PIP improved the phases by 20°.

Discussion

The PIP uses a MLA systematically to test alternative phases to improve DMM or CEDA results. During each cycle of the procedure an electron density map is calculated. This map is subjected to a DMM. From the modified map new phases are calculated by inverse Fourier transforms. These two steps give shifts for improving agreements between the density-modified and the starting electron density. These phases are altered to introduce some 'uphill steps' or to provide additional options during a MLA to change phases. The procedure electron density → DMM → phase change → electron density is iterated several times. Success of this procedure to improve phases depends on the ability of the DMM to correct non-beneficial changes made in the phase-change step. Different phase-change steps are suggested for the two symmetry cases. For centrosymmetric space groups, phases can be changed only by a large value, *i.e.* 180°, and hence, to reduce the mean phase change per cycle, phases of only a few selected reflections are changed. For a non-centrosymmetric case, phases of all reflections are changed by a small value using a function of the phase change suggested by the previous cycle. A selection criterion similar to that of the centrosymmetric case could have been used also in the non-centrosymmetric case. A more general form of such a criterion may include resolution as well as the magnitude of the structure factors, and phase shifts based on statistical considerations (Sim, 1959; Woolfson, 1956) in the phase-change step.

The test calculations were chosen to study the PIP incorporating a very simple DMM. This step used only the non-negativity rule of direct methods, though it would have been possible to apply other restraints [see the reviews of Podjarny, Bhat & Zwick (1987), Tulinsky (1985) and Wang (1985)] as well.

The parameters used in our calculations may not be optimal. Systematic attempts were not made to vary these parameters in a search for better results. The determination of the crystal structures of two

small molecules starting from the phases obtained by the CEDA, together with the results from the PIP illustrate *ab initio* phase determination and subsequent phase refinement. These were obtained using restraints currently employed in DMM. The results are encouraging, although the usefulness of the proposed method in routine structure determination requires further thought and investigation.

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Space Groups Rare for Organic Structures. II. Analysis by Arithmetic Crystal Class

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Abstract

Twelve arithmetic crystal classes contain only one space-group type; others contain various numbers up to sixteen ($mmmP$ and $4/mmmP$). In the multimember classes the number N_{sg} of substances having a particular space-group type can be modelled quantitatively (unweighted $R_2 \gtrsim 0.05$) by

$$N_{sg} = A_{cc} \exp \{\sum (X_i)_{cc}[x_i]_{sg}\},$$

where A_{cc} is a normalizing constant for the arithmetic crystal class, $[x_i]_{sg}$ is the number of symmetry elements $[2, 2_1, m, a, b, c, d, n, 3, \bar{3}, 3_1, 3_2, 4, \ldots]$ of type x_i in the unit cell of the space-group type, $(X_i)_{cc}$ is a parameter characteristic of the arithmetic crystal class and the symmetry element, and the summation is over all the elements x_i that are to be considered. In many cases the parameters X_i are equal, within their estimated standard deviations, throughout a cohort larger than the arithmetic crystal class (such as a geometric crystal class or an entire crystal system). The above equation can then be applied to the larger cohort, with arithmetic crystal class included as an additional 'symmetry element' in the sum in the exponent. There is at present no theory to account for the different popularities of different arithmetic crystal classes.

1. Introduction

The space-group type $P2_1/c$ accounts for about one-third of all known molecular organic structures, whereas the space-group type $P2/m$ has no certain example. Why? Ultimately the space group of a crystal of a particular substance is determined by the minimum (or a local minimum) of the thermodynamic potential (Gibbs free energy) of the van der Waals and other forces, but a very simple model goes a long way towards 'explaining' the relative frequency of the