

Even though the calculated values for the lattice energy could not be directly compared with the experimental ones because of the limitations of the approach used, it can be illustrative to compare the values obtained in the different runs when the hydrogen bonds are omitted. When the electrostatic term is not included, values of the lattice energy for rigid molecules (no subrotations) are in the range 70–100 kJ mol<sup>-1</sup>, whereas when molecular subrotations are relaxed lattice-energy values are in the range 100–160 kJ mol<sup>-1</sup>. For runs including the electrostatic term, the lattice energy is in the range 100–170 kJ mol<sup>-1</sup> when rigid molecules are considered and 130–200 kJ mol<sup>-1</sup> when molecular subrotations are relaxed. So, an unexpectedly large Coulombic contribution to the lattice energy, ranging from 30 to 60% of the total estimated lattice energy, is found. In our opinion, this high contribution is a qualitatively significant feature, in spite of the approach and the potential parameters used and the method of estimating effective atomic charges. Generally, it was tacitly assumed that the Coulombic contribution to the lattice energy is negligible but some previous results indicate that this assumption is not valid in all hydrocarbon crystals, reaching a value of 29% of the total lattice energy of benzene and up to 59% in other cases (Williams & Cox, 1984).

If the Coulombic contribution to the lattice energy seems to be significant for these crystals, it is true, however, that the inclusion of the electrostatic term does not significantly affect the values of the optimized structural parameters. The overall fit between the theoretical configuration and experimental structure is, in general, slightly more satisfactory for runs including the electrostatic contribution, but conclusive general results on the effects of its inclusion on molecular rotation and translation would be difficult to establish.

Within the limits of the approach and the potential functions used, we think that the reported results clearly indicate that the electrostatic term makes an important contribution to the lattice energy of these compounds but has little effect on the optimized structural parameters. A major limitation for accuracy in these calculations, in our opinion, is the description of the hydrogen bonds, but further improvements should be possible by defining a realistic potential function for these interactions.

#### References

- ALLINGER, N. L. & YUH, Y. (1980). Quantum Chemistry Program Exchange, **12**, 395. Indiana Univ.  
 BERTAUT, F. (1952). *J. Phys.* **13**, 499–505.  
 BURGOS, E. & BONADEO, H. (1977). *Chem. Phys. Lett.* **49**, 475–478.  
 CONDE, A., BERNIER, F. & MÁRQUEZ, R. (1980). *Acta Cryst.* **B36**, 3048–3052.  
 CONDE, C. F., MILLÁN, M., CONDE, A. & MÁRQUEZ, R. (1985a). *Acta Cryst.* **C41**, 277–280.  
 CONDE, C. F., MILLÁN, M., CONDE, A. & MÁRQUEZ, R. (1985b). *Acta Cryst.* **C41**, 1658–1662.  
 ESTRADA, M. D., CONDE, A. & MÁRQUEZ, R. (1983). *Acta Cryst.* **C39**, 1418–1421.  
 ESTRADA, M. D., CONDE, A. & MÁRQUEZ, R. (1984). *Acta Cryst.* **C40**, 898–901.  
 ESTRADA, M. D., CONDE, A. & MÁRQUEZ, R. (1986). *Acta Cryst.* **C42**, 454–457.  
 KITAIGORODSKY, A. I. (1973). *Molecular Crystals and Molecules*. New York: Academic Press.  
 MASON, A. & KREEVOY, M. M. (1955). *J. Am. Chem. Soc.* **77**, 5808–5814.  
 MIRSKAYA, K. V. (1973). *Tetrahedron*, **29**, 679–682.  
 MIRSKAYA, K. V. (1976). *Acta Cryst.* **A32**, 199–207.  
 RINALDI, R. P. & PAWLEY, G. S. (1973). *Nuovo Cimento B*, **16**, 55–62.  
 SKORCZYCK, R. (1976). *Acta Cryst.* **A32**, 447–452.  
 VEGA, R., HERNÁNDEZ-MONTIS, V. & LÓPEZ-CASTRO, A. (1976). *Acta Cryst.* **B32**, 1363–1366.  
 WILLIAMS, D. E. (1971). *Acta Cryst.* **A27**, 452–455.  
 WILLIAMS, D. E. (1972). *Acta Cryst.* **A28**, 629–635.  
 WILLIAMS, D. E. (1974). *Acta Cryst.* **A30**, 71–77.  
 WILLIAMS, D. E. & COX, S. R. (1984). *Acta Cryst.* **B40**, 404–417.

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## Structure and Charge Density of the 1:1 Complex of Thiourea with Parabanic Acid at 298 K

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#### Abstract

The crystal structure of the 1:1 complex of thiourea (CH<sub>4</sub>N<sub>2</sub>S) and parabanic acid (C<sub>3</sub>H<sub>2</sub>N<sub>2</sub>O<sub>3</sub>) at 298 K has been redetermined from X-ray intensity data (Mo K $\alpha$ , sin  $\theta/\lambda < 0.9 \text{ \AA}^{-1}$ ). The space group is

$P2_1/m$  (not  $P2_1$  as previously reported) with two molecules of each kind in the unit cell. Crystal data [ $M_r = 142.18$ ,  $F(000) = 148$ ] are:  $a = 10.874$  (4),  $b = 6.152$  (5),  $c = 6.161$  (3)  $\text{\AA}$ ,  $\beta = 114.39$  (3) $^\circ$  at 93 K;  $a = 10.893$  (5),  $b = 6.229$  (6),  $c = 6.160$  (3)  $\text{\AA}$ ,  $\beta = 114.31$  (4) $^\circ$  at 213 K;  $a = 10.916$  (2),  $b = 6.292$  (2),

$c = 6.158(2) \text{ \AA}$ ,  $\beta = 114.26(2)^\circ$  at 298 K. There are differences from the room-temperature constants previously reported:  $a = 10.984(19)$ ,  $b = 6.298(3)$ ,  $c = 6.238(12) \text{ \AA}$ ,  $\beta = 115.57(2)^\circ$ . The molecules form H-bonded planar sheets parallel to (010), separated by  $b/2 = 3.15 \text{ \AA}$ . Sheets stack with little molecular overlap, except that the S of thiourea is near the midpoint of the C–C bond of parabanic acid molecules in adjacent sheets. Analysis of the charge density assuming Stewart's rigid pseudoatom model shows no significant charge transfer between molecules. Molecular dipoles in one sheet are almost parallel with each other and are antiparallel to those in adjacent sheets. Mapping of the electrostatic potential and total electron density for atoms at rest shows similar features for the H-bonding interactions N–H $\cdots$ S and N–H $\cdots$ O. Comparison with the crystal structures of parabanic acid and thiourea shows the charge-density distribution to be similar for parabanic acid, but different, particularly at the S atom, for thiourea. These differences may not be real since results from the crystal structure of thiourea lack self-consistency.

### Introduction

The crystal structure of the 1:1 complex of thiourea and parabanic acid (TUPA, Figs. 1, 2) was first determined from X-ray (Cu  $K\alpha$ ) film data at room temperature by Colman & Medlin (1970). The space group was reported to be  $P2_1$  with all molecules lying in slightly puckered H-bonded sheets parallel to (010) and separated by  $b/2$  ( $3.15 \text{ \AA}$ ). The sheets stack such that the S atom of thiourea in one sheet is near the midpoint of the C–C bond in parabanic acid molecules of adjacent sheets (Fig. 2).<sup>\*</sup> In this region of overlap, Fourier syntheses  $\rho_o$  and  $(\rho_o - \rho_c)$  showed peaks of  $3.3$  and  $0.5 e \text{ \AA}^{-3}$  respectively at about  $0.8 \text{ \AA}$

<sup>\*</sup> Closest intermolecular distances S $\cdots$ C are  $3.23, 3.31(3) \text{ \AA}$ .

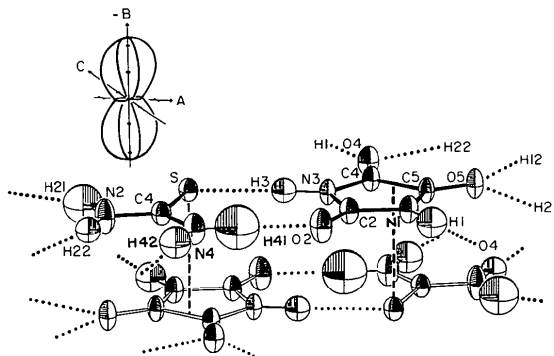


Fig. 1. Complex of thiourea and parabanic acid showing atomic nomenclature. Thermal-vibration ellipsoids are drawn at the 50% probability level (Johnson, 1976). Intermolecular interactions are drawn as dotted lines for H bonds and as broken lines for close S-atom distances. A polar graph of the mean thermal expansion coefficients between 93 and 298 K is drawn in the upper left part; each unit represents  $5 \times 10^{-5} \text{ K}^{-1}$ .

from the parabanic acid molecules. These features were of interest to us because of recent studies of the charge-density distribution in related crystal structures, including parabanic acid at 298 K (Craven & McMullan, 1979) and at 123 K (He, Swaminathan, Craven & McMullan, 1986), urea (Swaminathan, Craven, Spackman & Stewart, 1984) and thiourea (Mullen, Heger & Treutmann, 1978; Mullen, 1982; Kutoglu, Scheringer, Meyer & Schweig, 1982).

We have redetermined the crystal structure of TUPA with greater accuracy using X-ray (Mo  $K\alpha$ ) diffractometer data collected at room temperature. The new results indicate that the space group is  $P2_1/m$  rather than  $P2_1$ , so that the sheets of H-bonded molecules are planar. The unusual features in the electron density described by Colman & Medlin (1970) are absent in Fourier syntheses assuming either of the space groups  $P2_1$  or  $P2_1/m$ . Nevertheless, it was decided to carry out a charge-density analysis with the new X-ray data. There are limitations imposed by data collection at elevated temperature and the lack of neutron data for determining atomic positional and thermal parameters. However, in this crystal structure, there are advantages that the charge distribution can be checked for internal consistency due to the non-crystallographic symmetry of both molecules. Also, comparisons are possible with closely related structures. The resulting conclusions with regard to the electrostatic properties of the molecules and their interactions have exceeded expectations. The nature of the N–H $\cdots$ S interaction is of special interest.

### Experimental

Crystals grown by slow evaporation of an aqueous solution containing thiourea in excess. Pale yellow prisms elongated on [010], exhibiting {102}, {010},  $\{\bar{2}01\}$ , with (010) cleavage. X-ray data at 293 K from crystal  $0.36 \times 0.52 \times 0.12 \text{ mm}$  mounted almost along  $b^*$ . Enraf–Nonius CAD-4 diffractometer, graphite-monochromated Mo  $K\alpha$  ( $\lambda = 0.7093 \text{ \AA}$ ) gave 4030 intensities ( $\sin \theta/\lambda < 0.90 \text{ \AA}^{-1}$ ), 2902 after averaging equivalents ( $R_{int} = \sum |F - \langle F \rangle| / \sum \langle F \rangle = 0.013$ ). Scan by  $\omega/2\theta$ , width  $\Delta 2\theta = (1.5 + 0.35 \tan \theta)^\circ$ , time  $< 120 \text{ s}$ . Absorption correction ( $\mu = 3.77 \text{ cm}^{-1}$ ) and mean crystal path lengths from Gaussian approximation

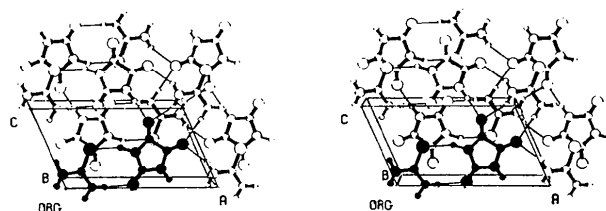


Fig. 2. Stereoview of the molecular packing. Spheres of decreasing size represent S, O, N, C and H. One molecular pair is shown with solid spheres. Thin lines between molecules represent H bonds.

(Busing & Levy, 1957). Corrected intensity 004 within 3% of mean value in azimuthal scan. Lattice parameters at 293 K from least-squares fit of  $\sin^2 \theta$  for 25 reflections  $14 < |\theta| < 26^\circ$ . Values at 213, 93 K from powder diffraction, Guinier-Simon camera ( $\lambda = 1.5418 \text{ \AA}$ ), films calibrated against Si ( $a_0 = 5.431 \text{ \AA}$  at 298 K).

Structure refinement by full-matrix least-squares computer program (Craven & Weber, 1982). Residual  $\sum w\Delta^2$  minimized where  $\Delta = |F_o| - |F_c|$  and  $w = \sigma^{-2}(F_o)$ , including all reflections.  $\sigma^2(F^2) = \sigma^2 + (0.02F^2)^2$  assumed with  $\sigma^2$  from counting statistics. Hence  $\sigma(F_o) = \sigma(F_o^2)/2|F_o|$  except for 651 weak reflections  $F^2 < \sigma(F^2)$  where  $\sigma(F_o) = \sigma(F^2)^{1/2}/2$  (Rees, 1976). Atomic scattering factors from Cromer & Waber (1965), except for H (Stewart, Davidson & Simpson, 1965). Refinement began in space group  $P2_1$ , positional and anisotropic thermal parameters from Colman & Medlin (1970), isotropic extinction parameter (type I crystal, Lorentzian mosaicity; Becker & Coppens, 1974), giving  $R = 0.063$ ,  $wR = 0.051$ ,  $S = 2.1$ .\* For non-H atoms,  $y$  coordinates all within  $3\sigma(y)$  of mean plane ( $y = \frac{1}{4}$  after transforming from arbitrary origin of Colman & Medlin, 1970). Space group  $P2_1/m$  (coordinates constrained,  $y = \frac{1}{4}$  strongly indicated by  $N(z)$  test (Howell, Phillips & Rogers, 1950). Refinement with  $P2_1/m$  gave  $R = 0.065$ ,  $wR = 0.054$ ,  $S = 2.2$ . Although  $P2_1$  preferred at 99.5% confidence level from  $R$ -factor ratio test (Hamilton, 1974), we assume  $P2_1/m$  because no physicochemical basis for slight sheet puckering could be found.

Further refinement was carried out with the pseudoatom model of Stewart (1976) to determine electronic charge-density distribution: pseudoatoms with neutral spherical cores corresponding to previous refinement; charge-density deformation terms, each the product of Slater-type radial and multipole angular function, with variable electron population parameter. Radial-function exponents assigned fixed standard values  $\alpha = 3.9, 4.5, 2.4, 4.1 \text{ bohr}^{-1}$  (the Bohr radius is  $52.92 \text{ pm}$ ) for N, O, H, S (Hehre, Stewart & Pople, 1969). For C,  $\alpha = 3.2 \text{ bohr}^{-1}$  as in parabanic acid (Craven & McMullan, 1979). For S, subsequent refinement gave  $\alpha = 5.9(1.6) \text{ bohr}^{-1}$ . Multipole expansion complete to octapole level (dipole for H). Explicit functions for deformations from Epstein, Ruble & Craven (1982); for S, same as P in Swaminathan & Craven (1984). Positional parameters for H fixed according to intramolecular geometry determined by neutron diffraction for the 1:1 complex of urea with parabanic acid (Weber, Ruble, Craven & McMullan, 1980). Because the mean-square amplitudes of atomic thermal vibrations were large in TUPA at 298 K ( $\sim 0.05 \text{ \AA}^2$  along  $b$ ), third-order ther-

mal parameters for non-H atoms were included. Possibly significant non-zero values for  $c_{jkl}$  found only for S in thiourea (see box, Fig. 3) and O(2) in parabanic acid.\* Refinement gave  $R = 0.048$ ,  $wR = 0.040$ ,  $S = 1.055$ , with final atomic positional and thermal parameters in Table 1,† and electron population parameters in Table 2. Extinction effects not severe [ $g = 0.033(6) \times 10^4 \text{ rad}^{-1}$ ]; reflection most affected is 020,  $(0.86|F_o|)$ . The finite difference Fourier map† has most significant residual density  $[0.40(16) \text{ e \AA}^{-3}]$  centered in the H...S bond.

Anisotropic thermal parameters for parabanic acid (omitting H atoms) were consistent with rigid-body motion. The agreement between  $U_{ij}$  values from Table 1 and the rigid-body model gave  $wR = 0.04$ , where weights  $w = \sigma^{-2}(U)$  were derived for each atom from the mean-square values of  $\sigma(U_{ij})$ . The rigid-body model is inappropriate for thiourea because non-H

\* For temperature factors of the form  $T = 1 - 4/3\pi^3 i \sum_l \sum_k \sum_j h_j h_k h_l c_{jkl}$ , the most significant values were  $c_{223} = -21(6) \times 10^{-7}$  for S and  $46(20) \times 10^{-7}$  for O(2).

† Difference Fourier synthesis maps and lists of bond lengths and angles, rigid-body thermal parameters, observed and calculated structure factors for TUPA and positional, thermal and electron population parameters for thiourea have been deposited with the British Library Document Supply Centre as Supplementary Publication No. SUP 43417 (34 pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 5 Abbey Square, Chester CH1 2HU, England.

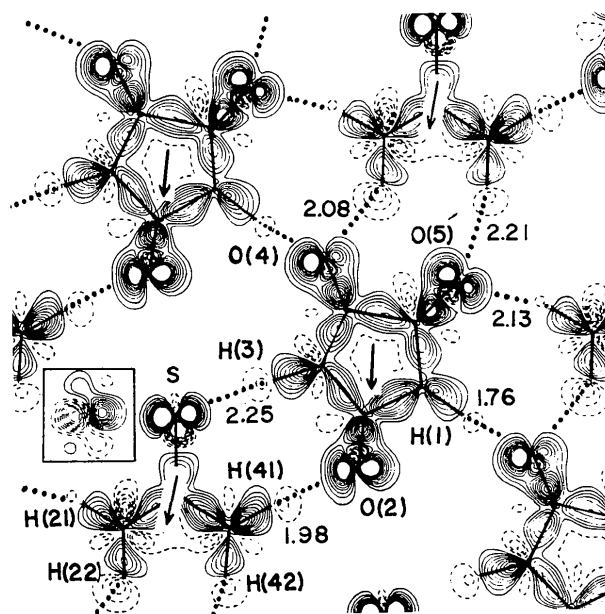


Fig. 3. Static deformation charge density for TUPA in the plane  $y = \frac{1}{4}$ . Contours are at intervals of  $0.1 \text{ e \AA}^{-3}$  with the zero contour omitted. The e.s.d. has a maximum value  $0.1 \text{ e \AA}^{-3}$  near the S atom. Directions of the molecular dipoles are shown by the heavy arrows at each molecular center. The inset near the S atom shows the skewed component of its probability distribution function due to third-order thermal vibration parameters. The inset is on twice the linear scale of the map. H bonds are represented by dotted lines with H...S or H...O distances in  $\text{\AA}$ .

$$* R = \frac{\sum |\Delta|}{\sum F_o}; wR = \left( \frac{\sum w\Delta^2}{\sum wF_o^2} \right)^{1/2}; \\ S = \left[ \frac{\sum w\Delta^2}{(m-n)} \right]^{1/2}.$$

Table 1. Atomic positional and anisotropic thermal parameters ( $\times 10^4$ )

Because of space-group symmetry, all atoms have  $y = \frac{1}{4}$  and  $U_{12} = U_{23} = 0$ . Values of  $U_{ij}$  ( $\text{\AA}^2$ ) are defined by the temperature-factor expression  $T = \exp(-2\pi^2 \sum_i \sum_j h_i h_j a_i^* a_j^* U_{ij})$ , or  $T = \exp(-8\pi^2 U_{11} \sin^2 \theta / \lambda^2)$  for H.

	x	z	$U_{11}$	$U_{22}$	$U_{33}$	$U_{13}$
N(1)	6850 (1)	2121 (2)	209 (5)	462 (8)	141 (4)	81 (4)
C(2)	5553 (1)	1975 (2)	179 (5)	412 (8)	145 (4)	36 (4)
O(2)	4537 (2)	159 (3)	216 (7)	739 (14)	202 (6)	11 (6)
N(3)	5622 (1)	4276 (2)	165 (5)	410 (8)	188 (4)	79 (4)
C(4)	6922 (1)	5890 (2)	178 (4)	374 (7)	125 (4)	66 (3)
O(4)	7373 (2)	8043 (3)	262 (6)	525 (10)	136 (5)	87 (5)
C(5)	7768 (1)	4430 (2)	163 (5)	401 (7)	155 (4)	70 (3)
O(5)	8979 (2)	5207 (3)	181 (6)	554 (11)	256 (6)	97 (5)
H(1)	6978	535	365 (119)			
H(3)	4732	4520	223 (92)			
S	2705 (3)	4628 (6)	185 (2)	349 (3)	199 (2)	49 (2)
C	1477 (1)	1810 (2)	188 (5)	339 (7)	203 (4)	43 (4)
N(4)	1730 (2)	-121 (3)	254 (7)	612 (12)	198 (5)	65 (5)
N(2)	199 (2)	1494 (4)	186 (6)	741 (15)	293 (7)	50 (5)
H(21)	-38	2916	672 (163)			
H(22)	-548	-149	473 (120)			
H(41)	2688	62	559 (146)			
H(42)	956	-1730	757 (137)			

atoms lie on a conic section (two intersecting straight lines; Johnson, 1970).

## Discussion

### (a) The crystal structure

In this crystal structure, the H-bonding and molecular packing arrangements (Figs. 1, 2) are quite different from the corresponding 1:1 urea complex (Weber *et al.*, 1980). Probably this is because TUPA cannot form the very short  $\text{NH}\cdots\text{O}$  interaction ( $\text{H}\cdots\text{O}$ , 1.61 Å) in which parabanic acid provides the N-H donor and urea the O acceptor. However, in TUPA there is a corresponding  $\text{NH}\cdots\text{S}$  interaction ( $\text{H}\cdots\text{S}$ , 2.25 Å) between the two molecules. This distance is shorter by 0.15 Å than is observed for any of the H bonds in the crystal structure of thiourea itself. In TUPA, the parabanic acid molecules are also H bonded to each other, with an  $\text{H}\cdots\text{O}$  distance (1.76 Å) similar to those in the crystal structure of parabanic acid (1.82, 1.89 Å; Craven & McMullan, 1979). In TUPA, the close packing of molecules within planar sheets brings about a short intermolecular  $\text{S}\cdots\text{O}(2)$  distance (3.16 Å) between molecules that are not directly H bonded.

In the revised space group  $P2_1/m$ , the distance between planar sheets of molecules is  $b/2 = 3.146(2)$  Å at 298 K. There is little overlap between molecules in adjacent sheets (Fig. 2), the only short interatomic distances being from the thiourea S atom to C(4) and C(5) in parabanic acid, 3.205, 3.278(2) Å. As might be expected from the weak interactions between layers, the crystal thermal expansion is greatest along **b** (inset, Fig. 1).

There are only marginally significant differences between bond lengths\* in TUPA and those determined by neutron diffraction in the crystal structures

of the component molecules. The largest differences involve C(4)-C(5), 0.014(3) Å and C(2)-O(2), 0.018(5) Å in parabanic acid. The longer C(2)-O(2) bond in TUPA is attributed to the H bonding of O(2), which does not occur in parabanic acid itself. Similar effects have been observed in related molecules (Craven, Cusatis, Gartland & Vizzini, 1973). The thiourea in TUPA is slightly distorted from ideal  $2mm$  point symmetry. Thus the S-C-N angles 123.1 and 119.3(1)° differ from each other and from the value [121.7(2)°] in thiourea. In TUPA, the smaller angle forms within the ring of the H-bonded cyclic dimer with parabanic acid.

### (b) Electrostatic properties

The net electronic charge on each pseudoatom is the negative value of the monopole population parameter ( $-p_v$ , Table 2). In order for the unit-cell contents to be electrically neutral,  $-\sum p_v$  for all atoms in the asymmetric unit should be zero. The value obtained [-0.26(36) e] is zero within experimental error. Furthermore, net charges on the parabanic acid and thiourea molecules are -0.52(22) and +0.26(26) e, indicating that any charge transfer in the crystal structure has at most a marginal significance in terms of experimental error. We find no significant difference in the charges on the C atoms in either molecule, and likewise for N, O and H.

Following Stewart (1972), molecular dipole moments have been calculated from the distribution of net charges and from the pseudoatom dipole population parameters. The value for thiourea [5.4(2.5) debye; 1 debye  $\equiv 3.34 \times 10^{-30}$  C m], agrees satisfactorily with the value 4.86 debye obtained for thiourea in dioxane solution (Kumler & Fohlen, 1942). A value for parabanic acid in solution is not available. However the dipole moment in TUPA [4.7(2.1) debye] agrees with the value calculated in the crystal structure of parabanic acid at 298 K [3.5(1.8) debye; He, Swaminathan, Craven & McMullan, 1986]. The molecular dipole moments in TUPA are in directions close to the bonds  $\text{S}\rightarrow\text{C}$  in thiourea and  $\text{C}(2)\rightarrow\text{O}(2)$  in parabanic acid (see Fig. 3). As a result, the molecular dipole moments are almost parallel for all molecules in a given sheet and are antiparallel to the dipole moments of molecules in the adjacent sheets.

The total electrostatic potential in the crystal structure of TUPA (Fig. 4a) has been calculated following Stewart (1982). This involves a Fourier synthesis with  $\Delta F/(\sin \theta/\lambda)^2$  as coefficients in order to obtain the contribution due to the deformation charge density. Here  $\Delta F = F_{\text{calc}} - F_{\text{IAM}}$ , where  $F_{\text{calc}}$  is the total structure factor for reflection  $hkl$ , and  $F_{\text{IAM}}$  is the contribution due to neutral spherical Hartree-Fock atoms. The contribution to the electrostatic potential due to each neutral spherical atom (including nucleus) is

\* See deposition footnote.

Table 2. *Electron population parameters* ( $\times 10^2$ )

Deformation terms are with respect to a molecular Cartesian axial system with  $x$  along the vector  $C(2) \rightarrow O(2)$  for parabanic acid ( $C \rightarrow S$  for thiourea) and  $z$  perpendicular to the molecular plane.\* Population parameters have normalized values (Epstein, Ruble & Craven, 1982).

	$p_0$	$d_1$	$d_2$	$d_3$	$q_1$	$q_2$	$q_3$	$q_4$	$q_5$	$o_1$	$o_2$	$o_3$	$o_4$	$o_5$	$o_6$	$o_7$
(a) Parabanic acid. Values for TUPA are above and those for parabanic acid at 298 K (He, Swaminathan, Craven & McMullan, 1986) are below.																
N(1)	11 (7)	9 (4)	7 (3)	0	-6 (3)	1 (4)	0	0	-12 (4)	-25 (4)	-4 (4)	0	0	1 (3)	1 (4)	0
	15 (2)	0 (1)	-1 (1)	-3 (1)	-5 (1)	5 (1)	1 (1)	1 (1)	-5 (1)	-23 (2)	-10 (2)	-3 (2)	2 (1)	3 (2)	1 (1)	7 (2)
C(2)	20 (10)	20 (4)	4 (4)	0	9 (3)	-1 (4)	0	0	-40 (4)	41 (4)	3 (4)	0	0	1 (3)	-4 (4)	0
	-1 (2)	13 (1)	2 (1)	5 (1)	15 (1)	-1 (1)	-1 (1)	0 (1)	-27 (2)	27 (2)	-4 (2)	-2 (2)	-3 (2)	5 (1)	-1 (1)	-1 (1)
O(2)	-4 (7)	9 (4)	5 (4)	0	-20 (4)	-9 (3)	0	0	-18 (4)	8 (3)	-7 (3)	0	0	-6 (4)	-10 (3)	0
	9 (2)	11 (1)	-1 (1)	2 (1)	-15 (1)	-1 (1)	3 (1)	0 (1)	-14 (1)	1 (1)	4 (3)	3 (1)	-2 (1)	2 (2)	-5 (2)	1 (1)
N(3)	22 (7)	5 (4)	-2 (3)	0	-9 (4)	-7 (3)	0	0	-5 (4)	-15 (3)	2 (4)	0	0	-2 (4)	3 (4)	0
	12 (2)	-3 (1)	6 (1)	-2 (1)	-1 (1)	-3 (1)	0 (1)	4 (1)	-5 (1)	-20 (2)	9 (2)	2 (2)	-1 (2)	-6 (2)	3 (2)	-2 (2)
C(4)	30 (8)	-9 (4)	-12 (3)	0	6 (4)	10 (4)	0	0	-38 (4)	16 (3)	-29 (4)	0	0	2 (4)	-2 (4)	0
	13 (2)	-6 (1)	-4 (1)	0 (1)	2 (1)	16 (1)	-4 (1)	-1 (1)	-29 (1)	8 (2)	-26 (2)	5 (2)	3 (1)	-1 (1)	-2 (1)	-4 (2)
O(4)	10 (6)	-1 (4)	-16 (3)	0	-9 (4)	-11 (3)	0	0	-15 (3)	-1 (4)	-2 (4)	0	0	-1 (4)	-6 (4)	0
	5 (2)	-9 (1)	-8 (1)	-6 (1)	-4 (1)	-7 (1)	-2 (1)	-7 (2)	-14 (2)	0 (2)	1 (2)	0 (2)	1 (1)	1 (1)	-3 (1)	5 (2)
C(5)	20 (9)	-5 (4)	15 (4)	0	1 (4)	-21 (3)	0	0	-36 (4)	13 (3)	39 (4)	0	0	3 (4)	-3 (3)	0
	12 (2)	-11 (1)	4 (1)	3 (1)	2 (1)	-15 (1)	1 (1)	-1 (1)	-25 (1)	14 (2)	26 (2)	4 (2)	-1 (2)	0 (1)	0 (2)	-5 (2)
O(5)	-1 (6)	-14 (4)	6 (3)	0	-13 (4)	14 (3)	0	0	-12 (3)	1 (4)	3 (4)	0	0	1 (3)	6 (3)	0
	5 (2)	-11 (1)	4 (1)	-4 (1)	-11 (1)	15 (1)	2 (1)	3 (1)	-11 (2)	-2 (2)	1 (2)	1 (2)	4 (2)	2 (1)	3 (2)	3 (2)
H(1)	-27 (7)	-4 (4)	-8 (4)	0												
	-19 (2)	-11 (1)	-14 (1)	1 (1)												
H(3)	-28 (6)	-7 (4)	12 (3)	0												
	-22 (1)	-6 (1)	18 (1)	2 (1)												
(b) Thiourea. Values for TUPA are above and those for molecules <i>A</i> and <i>B</i> in the crystal structure of thiourea are below.																
S	6 (8)	13 (6)	-5 (6)	0	-22 (3)	5 (3)	0	0	-13 (3)	-20 (6)	-1 (6)	0	0	14 (6)	-8 (6)	0
S(A)	22 (8)	-1 (6)	0	-22 (6)	-11 (4)	0	-1 (4)	0	-15 (4)	9 (3)	0	24 (3)	0	-4 (3)	0	22 (3)
S(B)	9 (8)	13 (5)	0	-20 (5)	0 (4)	0	1 (4)	0	3 (3)	-16 (3)	0	23 (3)	0	-13 (3)	0	12 (3)
C	-2 (9)	9 (3)	0 (4)	0	-3 (3)	-1 (3)	0	0	-25 (3)	26 (3)	7 (3)	0	0	0 (4)	3 (4)	0
C(A)	23 (9)	-2 (4)	0	-25 (4)	1 (3)	0	-5 (3)	0	-36 (3)	31 (2)	0	8 (3)	0	-4 (3)	0	7 (3)
C(B)	-19 (10)	5 (4)	0	-18 (4)	2 (3)	0	1 (3)	0	-16 (3)	14 (3)	0	3 (3)	0	3 (3)	0	14 (3)
N(4)	17 (9)	-1 (5)	-5 (3)	0	-7 (5)	-2 (5)	0	0	5 (4)	-23 (4)	-7 (4)	0	0	0 (5)	4 (4)	0
N(2)	4 (10)	9 (5)	7 (5)	0	-6 (5)	1 (5)	0	0	-12 (6)	-25 (4)	-4 (4)	0	0	1 (5)	1 (4)	0
N(A)	27 (10)	5 (3)	10 (3)	-13 (3)	-4 (3)	12 (3)	-3 (3)	-5 (3)	-8 (3)	-13 (2)	-5 (3)	6 (3)	-3 (3)	0 (3)	-9 (3)	5 (3)
N(B)	33 (10)	4 (3)	-7 (2)	-13 (2)	5 (2)	1 (3)	5 (2)	0 (3)	-3 (2)	-24 (2)	-2 (2)	7 (3)	-1 (3)	11 (2)	-7 (2)	0 (3)
H(21)	-18 (9)	-5 (5)	7 (5)	0												
H(41)	-15 (9)	-1 (4)	-15 (5)	0												
H(1A)	-10 (6)	15 (2)	3 (3)	5 (2)												
H(1B)	-27 (6)	9 (2)	-7 (2)	-1 (3)												
H(22)	-20 (9)	20 (5)	9 (4)	0												
H(42)	2 (9)	14 (5)	-3 (4)	0												
H(2A)	-28 (5)	-5 (2)	3 (2)	7 (2)												
H(2B)	-8 (6)	-3 (2)	-13 (2)	-2 (3)												

\* Inspection of the angular functions (Epstein *et al.*, 1982) shows that for idealized molecular symmetry  $2mm$ , atoms lying on the twofold axis should have  $d_2 = d_3 = q_2 = q_3 = q_4 = o_2 = o_3 = o_4 = o_6 = o_7 = 0$ . Other atoms should have  $d_3 = q_3 = q_4 = o_3 = o_4 = o_7 = 0$ . For atoms such as C(4) and C(5), which would become symmetry-related, corresponding values for  $d_1$ ,  $q_1$ ,  $q_5$ ,  $o_1$  and  $o_5$  should be equal, and for  $d_2$ ,  $q_2$ ,  $o_2$  and  $o_6$  should be equal and opposite in sign.

added separately as a radially symmetric component derived from the wave functions of Clementi (1965), together with an inner potential term which is analogous to  $F(000)$  in the Fourier series. The regions of negative potential are those which are low in energy for a unit positive charge as a test probe. Negative regions surround the molecules, the minimum value  $[-0.55 (7) e \text{ \AA}^{-1}]$  being in the molecular plane near the lone-pair region of O(4). In Fig. 4(a), the negative regions are seen as flattened basins bounded by steep walls except for ridges lying along the H bonds. The most prominent ridges are those which involve the two parabanic acid H atoms. One of these forms the  $N-H \cdots O$  interaction with the shortest  $H \cdots O$  distance (1.76 \text{ \AA}) in the crystal structure. The other forms the  $N-H \cdots S$  interaction ( $H \cdots S$ , 2.25 \text{ \AA}). Thus the map of electrostatic potential emphasizes the similarity of these two interactions, although chemically and geometrically, they are different. It should be noted (Fig. 4b) that a broad low ridge in electrostatic potential occurs between molecular layers in the region of the

short distances between S and the C(4)–C(5) bond.

The total charge density has been mapped for pseudoatoms at rest (Fig. 5). This was done, not as a Fourier series, but rather as a crystal-space summation of charge density for neutral spherical atoms derived from Clementi (1965), together with deformation terms from the pseudoatom model (Table 2). Calculations were for a group of four coplanar pairs of parabanic acid and thiourea molecules. The map is contoured at low levels of electron density in order to show details of the intermolecular regions. As in Fig. 4, the total electron density along each H bond involves a low ridge with a saddle point close to the  $H \cdots O$  or  $H \cdots S$  line of centers. In the formalism of Bader, Nguyen-Dang & Tal (1981), these saddle points are (3, -1) critical points indicating weak bonding interactions. It is interesting that in the two H bonds of the cyclic parabanic acid–thiourea dimer, the electron density at the saddle points is very similar ( $0.15 e \text{ \AA}^{-3}$ ) although the  $H \cdots S$  and  $H \cdots O$  distances differ by 0.27 \text{ \AA}.

(c) Comparisons with the charge density in other crystal structures

Electron population parameters for parabanic acid in TUPA are in good agreement with those from the crystal structure of parabanic acid at 123 and 298 K (He, Swaminathan, Craven & McMullan, 1986).<sup>\*</sup> This can be seen in Table 2(a), where parameters from both structures are referred to the molecular axial system, and in Figs. 3 and 6(c) which show the charge deformation density for parabanic acid in both crystal structures. There is close conformity to the  $2mm$  symmetry of the isolated molecule.

<sup>\*</sup> Results from X-ray and neutron diffraction data at 123 K have led to new refinements based on the 298 K data of Craven & McMullan (1979). It is notable that, with the revised electron population parameters (Table 2a), the lone-pair lobes are found to lie close to the molecular plane.

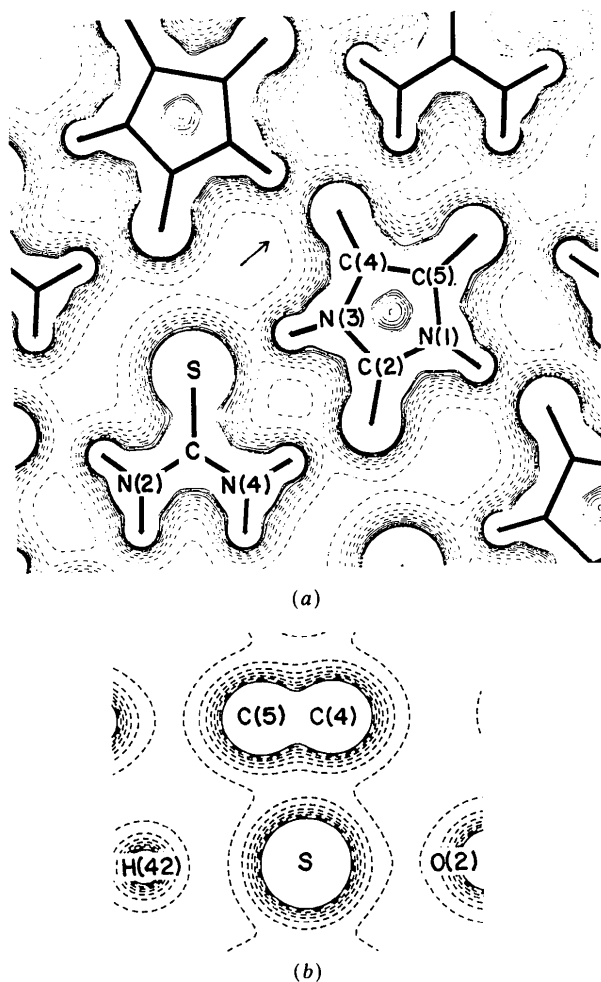


Fig. 4. Total electrostatic potential in the crystal structure of TUPA. Contours are at intervals of  $0.1 \text{ e } \text{Å}^{-1}$  ( $1 \text{ e } \text{Å}^{-1} = 1390 \text{ kJ mol}^{-1}$ ) with negative contours dotted. Negative regions are of low energy for a unit positive charge as probe. The position of the minimum ( $-0.55 \text{ e } \text{Å}^{-1}$ ) is indicated by the arrow. (a) The plane  $y = \frac{1}{4}$ , and (b) the plane through S and the  $b$  axis, parallel to the C(4)–C(5) bond.

The static deformation charge density for thiourea in TUPA (Fig. 3) and in the crystal structure of thiourea (Mullen, 1982; Kutoglu *et al.*, 1982) show notable differences. Thus the region near the S atom is almost featureless for both molecules *A* and *B* in the crystal structure of thiourea. To enable more direct comparisons to be made, we have carried out new refinements based on the X-ray data for thiourea collected at 123 K by Mullen & Hellner (1978) and subsequently revised by Kutoglu *et al.* (1982). We assumed the same pseudoatom structure model that was used for TUPA. Fixed positional parameters for H atoms were those determined by Elcombe & Taylor (1968) from neutron diffraction. The refinement converged with  $R = 0.014$ ,  $wR = 0.023$  and  $S = 0.543$ ,<sup>\*</sup> giving final values for positional and anisotropic thermal parameters<sup>†</sup> in very good agreement with those of Kutoglu *et al.* (1982). With our model, pronounced features occur in the static deformation charge density near the S atom in both molecules *A* and *B* (Figs. 6a,b). However, the electron population parameters (Table 2b), show the persistence of significant differences. The population parameters for thiourea in TUPA have values conforming reasonably well to an idealized molecular symmetry  $2mm$ . Those for molecules *A* and *B* in thiourea do not.<sup>‡</sup> The

<sup>\*</sup> Corresponding values from Kutoglu *et al.* (1982) are  $R = 0.0125$ ,  $wR = 0.0179$ ,  $S = 0.41$ . Since  $S < 1$ , it appears that  $\sigma(F)$  is overestimated.

<sup>†</sup> See deposition footnote.

<sup>‡</sup> The similarity in deformation density between molecule *A* (Fig. 6a) and thiourea in TUPA (Fig. 3) does not extend beyond the plane of the molecule.

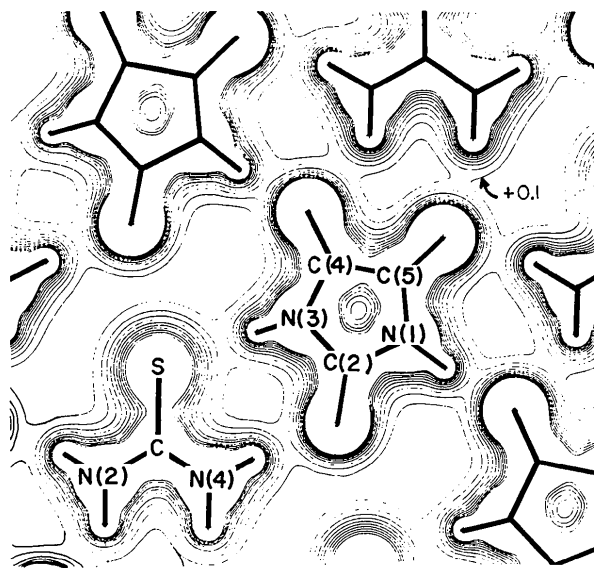


Fig. 5. Total charge density in TUPA ( $y = \frac{1}{4}$ ) for pseudoatoms at rest. Contours at intervals of  $0.1 \text{ e } \text{Å}^{-3}$  beginning at  $0.1 \text{ e } \text{Å}^{-3}$ . Calculations are for a finite group of four coplanar molecular pairs, and thus lack strict crystallographic periodicity.

departures from  $2mm$  symmetry are unexpectedly large, but may possibly be real since both molecules *A* and *B* have a similar crystal environment with an H-bonding arrangement which departs from  $2mm$  symmetry. For the same reason, significant differences between molecules *A* and *B* are unlikely to be real. As noted in the previous analyses (Mullen, 1982; Kutoglu *et al.*, 1982), these inconsistencies are most pronounced for the S atoms. The lone-pair lobes at S in molecule *A* (Fig. 6a) resemble those of thiourea in TUPA and also the ureide carbonyl atom O(2) in

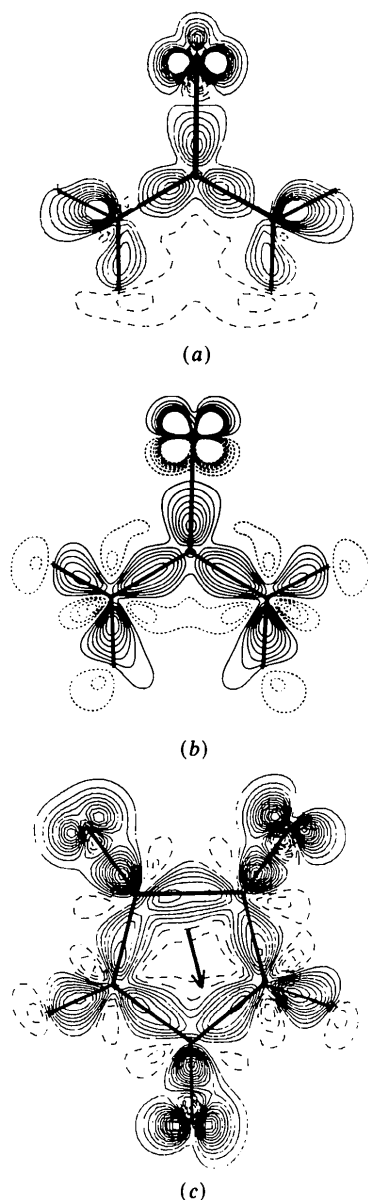


Fig. 6. Static deformation density in related crystal structures, derived from the electron population parameters in Table 2 (contours as in Fig. 3). (a) Thiourea at 123 K, molecule *A*. (b) Thiourea at 123 K, molecule *B*. (c) Parabanic acid at 298 K (He *et al.*, 1986).

TUPA (Fig. 3), primarily because of similar populations for the quadrupole deformation terms  $q_1$  and  $q_5$ . For molecule *B*, these populations are essentially zero. *Ab initio* quantum-mechanical calculations for thiourea (Kutoglu *et al.*, 1982) show significant deformation density very close to the S nucleus. In the experimental studies, none of the assumed models takes such features into account. In TUPA at 298 K, sharp features occur near the S nucleus. As in the case of the O atoms of parabanic acid at 298 K (He *et al.*, 1986), they have been interpreted as an antisymmetric component in the probability density function for thermal vibration (see box, Fig. 3).

Because of the internal inconsistencies in the charge density of molecules *A* and *B*, new low-temperature high-resolution X-ray data are needed for the crystal structure of thiourea. We plan no further work on TUPA.

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#### References

- BADER, R. F. W., NGUYEN-DANG, T. T. & TAL, Y. (1981). *Rep. Prog. Phys.* **44**, 894-948.
- BECKER, P. J. & COPPENS, P. (1974). *Acta Cryst.* **A30**, 129-147.
- BUSING, W. R. & LEVY, H. A. (1957). *Acta Cryst.* **10**, 180-182.
- CLEMENTI, E. (1965). *IBM J. Res. Dev. Suppl.* **9**, 2.
- COLMAN, P. M. & MEDLIN, E. H. (1970). *Acta Cryst.* **B26**, 1553-1559.
- CRAVEN, B. M., CUSATIS, C., GARTLAND, G. L. & VIZZINI, E. A. (1973). *J. Mol. Struct.* **16**, 331-342.
- CRAVEN, B. M. & MCMULLAN, R. K. (1979). *Acta Cryst.* **B35**, 935-945.
- CRAVEN, B. M. & WEBER, H. P. (1982). *The POP Least-Squares Refinement Procedure*. Tech. Rep. CC-74. Crystallography Department, Univ. of Pittsburgh.
- CROMER, D. T. & WABER, J. T. (1965). *Acta Cryst.* **18**, 104-109.
- ELCOMBE, M. M. & TAYLOR, J. C. (1968). *Acta Cryst.* **A24**, 410-420.
- EPSTEIN, J., RUBLE, J. R. & CRAVEN, B. M. (1982). *Acta Cryst.* **B38**, 140-149.
- HAMILTON, W. C. (1974). *International Tables for X-ray Crystallography*, Vol. IV, pp. 285-310. Birmingham: Kynoch Press. (Present distributor D. Reidel, Dordrecht.)
- HE, X., SWAMINATHAN, S., CRAVEN, B. M. & MCMULLAN, R. K. (1986). Unpublished.
- HEHRE, W. J., STEWART, R. F. & POPLE, J. A. (1969). *J. Chem. Phys.* **51**, 2657-2664.
- HOWELL, E. R., PHILLIPS, D. C. & ROGERS, D. (1950). *Acta Cryst.* **3**, 210-214.
- JOHNSON, C. K. (1970). In *Crystallographic Computing*, edited by F. R. AHMED, p. 214. Munksgaard: Copenhagen.
- JOHNSON, C. K. (1976). *ORTEP II*. Report ORNL-5138. Oak Ridge National Laboratory, Tennessee.
- KUMLER, W. D. & FOHLEN, G. M. (1942). *J. Am. Chem. Soc.* **64**, 1944-1946.
- KUTOGLU, A., SCHERINGER, C., MEYER, H. & SCHWEIG, A. (1982). *Acta Cryst.* **B38**, 2626-2632.
- MULLEN, D. (1982). *Acta Cryst.* **B38**, 2620-2625.
- MULLEN, D., HEGER, G. & TREUTMANN, W. (1978). *Z. Kristallogr.* **148**, 95-100.
- MULLEN, D. & HELLNER, E. (1978). *Acta Cryst.* **B34**, 2789-2794.

- REES, B. (1976). *Acta Cryst.* **A32**, 483-488.  
 STEWART, R. F. (1972). *J. Chem. Phys.* **57**, 1664-1668.  
 STEWART, R. F. (1976). *Acta Cryst.* **A32**, 565-574.  
 STEWART, R. F. (1982). *God. Jugosl. Cent. Kristalogr.* **17**, 1-24.  
 STEWART, R. F., DAVIDSON, E. R. & SIMPSON, W. T. (1965). *J. Chem. Phys.* **42**, 3175-3187.

- SWAMINATHAN, S. & CRAVEN, B. M. (1984). *Acta Cryst.* **B40**, 511-518.  
 SWAMINATHAN, S., CRAVEN, B. M., SPACKMAN, M. A. & STEWART, R. F. (1984). *Acta Cryst.* **B40**, 398-404.  
 WEBER, H.-P., RUBLE, J. R., CRAVEN, B. M. & McMULLAN, R. K. (1980). *Acta Cryst.* **B36**, 1121-1126.

*Acta Cryst.* (1987). **B43**, 209-212

## Location of Iron and Sulfur Atoms in Myohemerythrin from Anomalous-Scattering Measurements

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### Abstract

Anomalous scattering due to the iron and sulfur atoms in myohemerythrin has provided a check on the iron-iron distance and the location of the sulfur atoms which is independent of our model obtained by restrained least-squares refinement. By anomalous-scattering techniques we find an iron-iron distance of 3.35 (5) Å which compares with a distance of 3.23 Å obtained from restrained least-squares refinement. We have also used anomalous-scattering information to confirm the placement of Cys35S $\gamma$ , which required a sequence change during the refinement, and to establish our tentative identification of two solvent molecules as sulfate ions. Methods used here might also help in locating metal centers and finding the sites of minor anomalous scatterers in other macromolecular structures.

### Introduction

The use of anomalous scattering has allowed a model-independent assessment of the location of iron and sulfur atoms in the structure of myohemerythrin, an oxygen-carrying protein from sipunculan worms. We have used Bijvoet-difference Patterson maps (Rossmann, 1961), refinement against 'large' observed Bijvoet differences (Hendrickson, Klippenstein & Ward, 1975), Bijvoet-difference Fourier maps (Kraut, 1968) and the 'imaginary' Fourier synthesis (Hendrickson & Sheriff, 1987) in order to investigate the structure of anomalous scatterers in myohemerythrin.

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### Bijvoet-difference Patterson maps

Myohemerythrin from *Themiste zostericola* crystallizes from 2.3 M (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> in space group *P*2<sub>1</sub>2<sub>1</sub>2<sub>1</sub> with *a* = 41.66, *b* = 80.17, *c* = 37.82 Å. The active center contains two iron atoms ligated directly to protein side chains. One iron is coordinated by three histidines (His 73, 77, 106) and the other by two (His 25, 54). There are three connecting groups between the two iron atoms - the carboxylates of Glu 58 and Asp 111 and an oxide ion, O<sub>2</sub><sup>2-</sup>. The remaining ligand is azide in the azidomet form of hemerythrin (Sheriff, Hendrickson & Smith, 1983; Stenkamp, Sieker, Jensen & Sanders-Loehr, 1981).

Rossmann (1961) showed that Patterson maps calculated with  $|\Delta F|^2$ , where  $\Delta F = (|F^+| - |F^-|)$  is the Bijvoet difference, would yield the interatomic vectors of the anomalous scatterers in a structure. In Fig. 1 we show the three Harker sections from the Bijvoet-difference Patterson map at a nominal resolution of 2.0 Å. The iron self-peaks are clearly resolved in all three sections. In the *u* = *a*/2 and *v* = *b*/2 sections an iron-atom cross vector is also apparent.

### Refinement of the iron positions

The Bijvoet difference can be calculated as follows for a single type of anomalous scatterer

$$\Delta F \approx -2\delta \sin(\psi - \varphi) \quad (1)$$

where the values are as defined in Fig. 1 of Hendrickson (1979):  $\Delta F$  is the Bijvoet difference  $|F(\mathbf{h})| - |F(-\mathbf{h})|$ ,  $\delta \exp(i\psi)$  is the structure-factor contribution from anomalous-scattering centers, and  $\varphi$  is the phase for the complete structure. As  $|\Delta F|$  becomes large the value of the sine factor must approach  $\pm 1$ , which reduces equation (1) to  $|\Delta F| = 2\delta$ , for large