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Crystal and Molecular Structure of 1,3,5-Tri-*tert*-butylbenzene

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The molecule of 1,3,5-tri-*tert*-butylbenzene, $C_{18}H_{30}$, $M_r = 246\cdot44$, crystallizes in the monoclinic space group $C2/c$ with $a = 10\cdot193$ (2), $b = 17\cdot650$ (3), $c = 20\cdot222$ (5) Å, $\beta = 90\cdot00$ (1)°, $Z = 8$, $U = 3638\cdot1$ Å^3, $D_x = 0\cdot90$ g cm^{-3}. The reflections obey extra rules regarding extinction and intensity. The structure was analyzed by using these rules and refined by the least-squares method to an R value of 0·087 for the observed reflections. The disordered *tert*-butyl model corresponds well to the analysis. The molecule is found to have the symmetry of the pseudo $\bar{6}m2$ point group. One methyl in every *tert*-butyl group lies on the aromatic plane.

Experimental

The crystals of 1,3,5-tri-*tert*-butylbenzene, $C_{18}H_{30}$, used for the X-ray study were grown by evaporation from a solution of alcohol at room temperature. The crystals are colorless, transparent oblique plates. The melting point of the purified material was 67 °C.

In order to measure the crystal parameters and the intensity data, the crystal had to be enclosed in a thin-walled glass capillary to avoid rapid sublimation.

The cell dimensions were measured by using a two-circle diffractometer (SG-5 type, Rigaku–Denki Co.) with Cu $K\alpha_1$ and Cu $K\alpha_2$ radiation. A crystal with a diameter of less than 0·1 mm was used for this purpose. The 2θ values of the reflections were obtained from the measurements on both sides of the incident beam by using a narrow receiving slit. The lattice parameters a and b are deduced from $kk0$, $k\bar{k}0$, $\bar{k}k0$, $\bar{k}\bar{k}0$, $0,2k,0$ and $0,2\bar{k},0$ reflections of large indices by mounting the crystal along the c axis. The parameters c and β were determined from similar measurements of the $h0l$ reflections by mounting the crystal along the b axis.

The crystal used for the intensity measurements had dimensions of approximately 0·3 × 0·3 × 0·4 mm. Intensity data were collected with unfiltered Cu K radiation from zero to fifth layer equi-inclination multi-film Weissenberg photographs about the a axis, zero to seventh layer photographs about the b axis and zero and first layer about the c axis. Photographs along the $[\bar{1}10]$ and $[\bar{3}\bar{1}0]$ directions were also collected. These directions are referred to hereafter as fictitious axes and written as a_f and b_f axes, respectively.

The intensities were estimated visually, corrected for Lorentz and polarization factors and brought to a common scale in the usual way (Takwale & Pant, 1971). About 900 independent reflection intensities were obtained. It is estimated that nearly 4000 independent reflections are available in the Cu K C-centered monoclinic tetarto-sphere. Since the observed reflections of all expected types extend to the limit of the sphere of reflection, 3100 unobserved reflections were considered too weak to be detected and some of them follow a sort of regularity in the reflection indices. Weissenberg photographs along the a axis are useful for understanding these additional rules.

Special rules of extinction and regularity of intensities

Extra rules of extinction are found in addition to the systematic absence of reflections, which show that the space group of the crystal is $C2/c$ or Cc. Such additional extinction rules have been reported by many authors (for example, Dunitz, 1964; Dornberger-Schiff, 1966). They pointed out that one has to take account of twin formation of the crystal with a special ratio of lattice parameters. However, in the present case the additional extinction rules are not due to twin formation. These are caused by the molecule having a disorder character with a symmetry higher than that permitted by its chemical structural formula and also caused by a particular shape and orientation and the position of the molecule in the cell.

The $F_o(hkl)$'s which should be observed in an ordinary C-centered structure are not found in the following conditions:

$$
\begin{aligned}
&\text{(i) } hkl\colon k = 3n_k,\ l = 2n_l + 1\\
&\text{(ii) } 0kl\colon k = 2n_k,\ l = 4n_l + 2 \text{ and } (k = 6n_k,\ l = 2n_l + 1)\\
&\text{(iii) } h0l\colon (h = 2n_h,\ l = 2n_l + 1)\\
&\text{(iv) } 00l\colon (l = 4n_l + 2) \text{ and } (l = 2n_l + 1)
\end{aligned}
\qquad (1)
$$

where n_h, n_k and n_l are integers and parentheses indicate that the condition enclosed is included in a more general condition. Exceptions are 022 and $13\bar{1}$ of weak intensity and they require large Lp corrections.

Regularity was also found for a pair of structure factors for all observed reflections:

$$|F_o(hkl)| = |F_o(hk\bar{l})| \quad (l\text{: even}). \tag{2}$$

The additional extinction rules (1) limit the positions and the shapes of the molecules. The rule (1i) indicates that the local symmetry of charge density must satisfy the relation

$$\rho(x,\tfrac{1}{6}+\Delta y,z)=\rho(x,\tfrac{1}{6}-\Delta y,z). \tag{3}$$

In addition, it is noticed that the equator photograph along the b_f axis shows the same pattern as that of the b axis, namely

$$|F_o(h0l)| = \left|F_o\left(\frac{-h}{2}\,\frac{3h}{2}\,l\right)\right|. \tag{4}$$

This is a useful relation. By combining with relation (3), it restricts the symmetry of the molecules to some of the trigonal $3m$, $\bar{3}m$ and the hexagonal $6mm$, $\bar{6}m2$ and $6/mmm$ point groups, and also the x coordinates of the centers of the molecules to $x_1=\frac{1}{6}$ and $x_2=-\frac{1}{6}$. Further, the point groups $\bar{6}m2$ and $6/mmm$ from among those satisfy the following relation

$$\begin{aligned}\rho(x_j,\tfrac{1}{6}+\Delta y,z_j+\Delta z)&=\rho(x_j,\tfrac{1}{6}-\Delta y,z_j+\Delta z)\\ &=\rho(x_j,\tfrac{1}{6}+\Delta y,z_j-\Delta z)=\rho(x_j,\tfrac{1}{6}-\Delta y,z_j-\Delta z)\\ &(j=1,2)\end{aligned} \tag{5}$$

where $z_1=\frac{1}{8}$, $z_2=\frac{3}{8}$ and x_j's are arbitrary values.

The equality (5) is the adoptable one of the relations deduced from the additional rules of extinction (1ii) and of intensity (2) when the crystal belongs to the space group Cc and $Z=8$. On the other hand, this equality is the only relation derived from these additional rules if the crystal belongs to the space group $C2/c$. In the present crystal, the former space group Cc requires two crystallographically independent molecules with the same orientation at $(\frac{1}{6},\frac{1}{6},\frac{1}{8})$ and $(-\frac{1}{6},\frac{1}{6},\frac{3}{8})$ whereas the latter $C2/c$ requires one at $(\frac{1}{6},\frac{1}{6},\frac{1}{8})$.

The simplest way for selecting the space group of the crystal and the shape of the molecule is to consider the detailed physical nature of the $F_o(h00)$'s. The resultant scattering amplitude in the $[h00]$ direction by two molecules containing one species of atoms is

$$F(h00)=A_{\max}(h00)\times\sum_{j=1,2}\exp\,[i\Phi_j(h00)],$$

$$\Phi_j(h00)=2\pi hx_j+\varphi(h00) \qquad (j=1,2) \tag{6}$$

where x_j $(j=1,2)$ represent the x coordinates of the centers of two molecules (Wilson, 1966). Notations $A_{\max}(h00)$, $\Phi_j(h00)$ and $\varphi(h00)$ represent the maximum resultant amplitude, the resultant phase angle of the jth molecule and that when $x_j=0$, respectively. Quantities corresponding to the present molecule are shown in Table 1. In the calculation of the table, the molecule is tentatively assumed to belong to the point group $\bar{6}m2$ with usual bond lengths and angles. One *tert*-butyl is also assumed to be parallel to the x axis and oriented to $-x$. Hereafter, the state of the point symmetry $\bar{6}m2$ and others of lower symmetries are referred to as the disordered and ordered states respectively.

The maximum resultant amplitude $A_{\max}(400)$ is not very small in comparison with $A_{\max}(200)$. In the space group Cc, the equation to reproduce the observed $F_o(400)=0$ in $F(400)$ is derived from (6)

$$\Phi_1(400)-\Phi_2(400)=(2n+1)\pi.$$

The relation $x_1-x_2=(2n+1)/8$ (n: integers) is inconsistent with that derived from (4). The equation in the case of $C2/c$ is

$$\Phi_1(400)=(n+\tfrac{1}{2})\pi \quad [\because \Phi_2(h00)=-\Phi_1(h00)]$$

which gives

$$x_1=0{\cdot}167+n/8 \quad (n\text{: integers}).$$

The two-dimensional Fourier map projected down the a_f axis along with the $F_o(\bar{k},k,\pm l)$'s determines the true position $n=0$, which is consistent with the coordinate obtained from (4). The more disordered molecular model $6/mmm$ is unreasonable because it has an inversion center $[\varphi'(h00)=\pi$ or $0]$.

Table 1. *The resultant maximum amplitudes, phase angles when the x coordinate of the center of the molecule is zero and observed structure factors in the [h00] direction for the molecule*

h	2	4	6	8
$A_{\max}(h00)$	44	20	1	2
$\varphi(h00)$	1·85	−2·62	–	–
$\lvert F_o(h00)\rvert$	very large	0	0	0

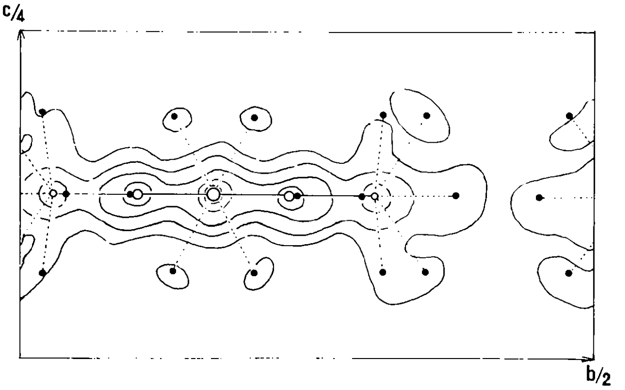

Fig. 1. Projection of the molecule along the a axis. The (100) projection of the two-dimensional charge-density map obtained from $F_o(0kl)$'s is also shown. In the figure, large and medium open circles indicate three and two carbon atoms aligned normal to the plane of projection, respectively. A small open circle indicates one atom and a small black circle a half-atom.

Refinement of the structure

The coordinates of the atoms of the molecule were obtained by two-dimensional Fourier maps projected along the a and a_f axes. In both projections, the charge-density maps of the molecule have the appearance of the pseudo $2mm$ symmetry and indicate the existence of half-methyls. Fig. 1 is the (100) projection map of charge density drawn from the $F_0(0kl)$'s. In Fig. 1, the final positions of the atoms are also shown.

The structure was first refined with the help of the atomic positions obtained from these Fourier syntheses for general reflections by using an overall temperature factor. Further refinements were carried out first with individual isotropic and later with anisotropic temperature factors. During these cycles a few thermal parameters were not varied because the symmetry of the molecule shows some components to be zero. A difference Fourier map synthesized after these cycles indicated that the molecular models of the ordered states are not correct in the present crystal. The number of possible ordered states of the molecule is eight (2 orientations for each 3 *tert*-butyls). The R value of the disordered state was 0·095 at this stage, whereas those of the ordered ones did not fall lower than 0·128. As the H atom positions in the butyls were not obtained even at the end of the refinement, an assumption was introduced in the last two cycles. All H atoms in butyls were placed around the half-carbon atoms. For the calculation, one and a half H atoms were statistically distributed into 12 segments and arranged on the circumference of a half-carbon, which is the trace of the H atoms in an ordinary rotating methyl radical. The R value of 0·087 was obtained finally. In the whole cycles, unit weight was given to all F_o's. The scattering factors were taken from *International Tables for X-ray Crystallography* (1968).

Table 2. *Final atomic parameters*

The e.s.d.'s of atomic parameters are given in parentheses and refer to the last significant figures. Atoms with an asterisk are the half-atoms.

	x	y	z
C(1)	0·0309 (9)	0·1664 (8)	0·1252 (5)
C(2)	0·0993 (12)	0·2351 (7)	0·1234 (7)
C(3)	0·2334 (11)	0·2351 (6)	0·1254 (7)
C(4)	0·3048 (9)	0·1677 (7)	0·1245 (5)
C(5)	0·2355 (11)	0·0980 (6)	0·1253 (7)
C(6)	0·0997 (11)	0·0979 (6)	0·1252 (6)
C(7)	−0·1210 (8)	0·1678 (7)	0·1262 (5)
C(8)	0·3108 (11)	0·3088 (7)	0·1252 (7)
C(9)	0·3092 (12)	0·0230 (7)	0·1247 (7)
C(10)*	−0·1752 (27)	0·2526 (14)	0·1260 (20)
C(11)*	−0·1726 (24)	0·2079 (22)	0·0644 (15)
C(12)*	−0·1686 (26)	0·1258 (21)	0·0645 (16)
C(13)*	−0·1682 (27)	0·0843 (15)	0·1263 (19)
C(14)*	−0·1729 (26)	0·1269 (19)	0·1866 (14)
C(15)*	−0·1717 (26)	0·2127 (19)	0·1860 (16)
C(16)*	0·4619 (26)	0·2910 (15)	0·1234 (19)
C(17)*	0·3999 (31)	0·3133 (17)	0·0620 (13)
C(18)*	0·2729 (27)	0·3552 (13)	0·0620 (12)
C(19)*	0·2120 (25)	0·3749 (15)	0·1239 (18)
C(20)*	0·2794 (31)	0·3555 (14)	0·1859 (14)
C(21)*	0·4067 (29)	0·3128 (13)	0·1871 (12)
C(22)*	0·2099 (26)	−0·0435 (15)	0·1261 (18)
C(23)*	0·2690 (29)	−0·0221 (15)	0·0621 (16)
C(24)*	0·3931 (26)	0·0182 (16)	0·0636 (13)
C(25)*	0·4532 (32)	0·0384 (14)	0·1210 (22)
C(26)*	0·3920 (31)	0·0214 (19)	0·1829 (18)
C(27)*	0·2690 (33)	−0·0242 (16)	0·1874 (15)
H(2)	0·0170 (128)	0·2813 (69)	0·1324 (74)
H(4)	0·4042 (78)	0·1653 (55)	0·1288 (41)
H(6)	0·1438 (87)	0·0481 (47)	0·1214 (46)

Discussion

The final atomic parameters along with their estimated standard deviations are given in Table 2.* Intramolecular distances and angles are listed in Table 3. Fig. 2 presents a drawing of the molecule and indicates the numbering scheme.

The average length of the aromatic bonds is 1·394 Å. The mean bond length between a C atom in an aromatic ring and the central C atom of a *tert*-butyl group, the bond C(1)–C(7) is an example, is 1·531 Å. These values agree within experimental error with those in other aromatic compounds. The mean value of the C–C bonds in the aliphatic groups is 1·549 Å. For the *tert*-butyl group, the number of methyl groups attached to the central C atom is thought to be six half-methyls and the butyl group seems to have a pseudo-hexad rotation axis. Two half-methyls in the opposite position are located in the benzene ring plane. The

Table 3. *Bond lengths* (Å) *and angles* (°)

Values for the *tert*-butyl groups are omitted, because the molecule is in a disordered state and such bond lengths and angles have little meaning. For numbering see Fig. 2.

C(1)–C(2)	1·401	C(1)–C(2)–C(3)	119·8
C(1)–C(6)	1·398	C(6)–C(1)–C(2)	119·9
C(1)–C(7)	1·549	C(2)–C(1)–C(7)	119·0
C(3)–C(4)	1·393	C(6)–C(1)–C(7)	121·1
C(3)–C(2)	1·367	C(3)–C(4)–C(5)	118·7
C(3)–C(8)	1·522	C(2)–C(3)–C(4)	121·5
C(5)–C(6)	1·383	C(4)–C(3)–C(8)	117·3
C(5)–C(4)	1·419	C(2)–C(3)–C(8)	121·1
C(5)–C(9)	1·522	C(5)–C(6)–C(1)	120·1
		C(4)–C(5)–C(6)	119·9
		C(6)–C(5)–C(9)	119·5
		C(4)–C(5)–C(9)	120·5

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* Lists of structure factors and anisotropic thermal parameters have been deposited with the British Library Lending Division as Supplementary Publication No. SUP 33828 (6 pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 5 Abbey Square, Chester CH1 2HU, England.

molecule has the appearance of pseudo $\bar{6}m2$ symmetry, although there are no crystallographic requirements.

The stacking form of the molecules in a unit cell has the aromatic plane exactly perpendicular to the c axis and four layers of molecules piled up along this axis. The position of the center of the molecule is $x = \frac{1}{8}(0{\cdot}0008)$, $y = \frac{1}{8}(0{\cdot}0004)$ and $z = \frac{1}{8}(0{\cdot}0004)$. The arrangement of the molecules projected on a plane perpendicular to the c axis is shown in Fig. 3. The nearest C—C distance between two half-methyls belonging to the adjacent molecules in the same (001) plane is 4·0 Å. The distance between half-methyls of one layer of molecules and those of the next layer is also 4·0 Å.

The principal r.m.s. displacements of the individual atomic vibration ellipsoids of each C atom in the benzene core are of the same order of magnitude for their three principal axes. The thermal vibration of half-carbon atoms in the *tert*-butyl groups occurs strongly towards the direction of the circumference of the ring composed of six half-methyls.

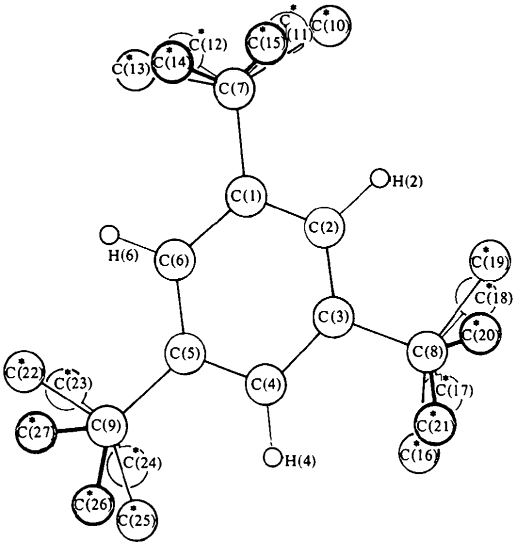

Fig. 2. A view of the molecule showing the numbering scheme. Atoms with an asterisk are the half-carbon atoms.

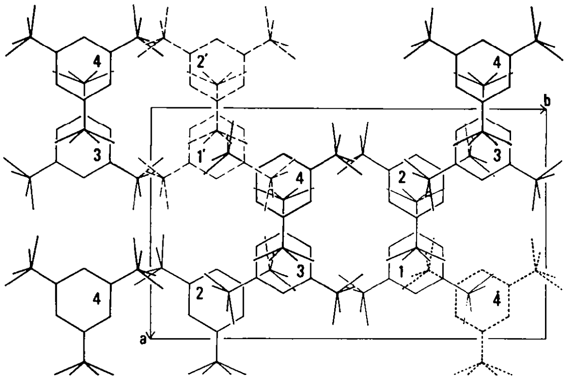

Fig. 3. Projection of the molecules in the unit cell along the c axis. Numbers 1, 2, 3 and 4 mean the first, second, third and fourth layers in a cell, respectively. The z coordinates are at $\frac{1}{8}$, $\frac{3}{8}$, $\frac{5}{8}$ and $\frac{7}{8}$.

It is expedient to treat the motion of the molecule as a rigid body (Cruickshank, 1956*a,b*, 1957*a,b*), because the highly symmetrical molecules are packed in the C-centered form and piled in layers. 12 independent parameters, including non-diagonal terms, are calculated by the least-squares method using 243 parameters (3 positional and 6 thermal for 9 carbon and 18 half-carbon atoms). The resulting matrices **T** and **Ω** are

$$\mathbf{T} = \begin{bmatrix} 0{\cdot}0567 & 0{\cdot}0026 & -0{\cdot}0024 \\ & 0{\cdot}0706 & -0{\cdot}0036 \\ & & 0{\cdot}0527 \end{bmatrix} \text{Å}^2$$

$$\mathbf{\Omega} = \begin{bmatrix} 16{\cdot}439 & -4{\cdot}285 & -0{\cdot}032 \\ & 12{\cdot}562 & 0{\cdot}885 \\ & & 10{\cdot}675 \end{bmatrix} \text{deg}^2.$$

To show the motion of the molecule more simply, let the molecule belong to the idealized hexagonal point group $\bar{6}m2$ and the internal axes belonging to the molecule, X, Y and Z, be taken parallel to the crystal axes x, y and z. Then the non-diagonal elements of the matrices **T** and **Ω** must be zero and the diagonal ones are written as

$$T_X = T_Y = 0{\cdot}06 \text{ Å}^2 \text{ and } T_Z = 0{\cdot}05 \text{ Å}^2$$
$$\Omega_X = \Omega_Y = 14{\cdot}5 \text{ deg}^2 \text{ and } \Omega_Z = 10{\cdot}7 \text{ deg}^2.$$

The amplitudes of the translational vibrations are of the same order for the three axes. The libration angle about the axis normal to the molecular plane is rather smaller than that parallel to it.

More detailed discussions on the disorder character of the molecule are impossible in the present study. However, further studies including measurements of heat capacity and absorption spectra should give information about the symmetry of the molecule. These experiments are now in progress.

The calculations were carried out on a FACOM M-190 computer at the Data Processing Center, Kyoto University. Programs for least-squares refinements Fourier syntheses and molecular motions as a rigid body were *RSFLS*-4, *RSSFR*-3 and *RSMV*-4 in *UNICS* (1967).

References

Cruickshank, D. W. J. (1956*a*). *Acta Cryst.* **9**, 754–756.
Cruickshank, D. W. J. (1956*b*). *Acta Cryst.* **9**, 915–923.
Cruickshank, D. W. J. (1957*a*). *Acta Cryst.* **10**, 470.
Cruickshank, D. W. J. (1957*b*). *Acta Cryst.* **10**, 504–508.

DORNBERGER-SCHIFF, K. (1966). *Acta Cryst.* **21**, 311–322.
DUNITZ, J. D. (1964). *Acta Cryst.* **17**, 1299–1304.
International Tables for X-ray Crystallography (1968). Vol. III. Birmingham: Kynoch Press.
TAKWALE, M. G. & PANT, L. M. (1971). *Acta Cryst.* B**27**, 1152–1158.
UNICS (1967). *The Universal Crystallographic Computing System*(I), edited by T. SAKURAI. Tokyo: The Crystallographic Society of Japan.
WILSON, H. R. (1966). *Diffraction of X-rays by Proteins, Nucleic Acids and Viruses*, p. 28. London: Edward Arnold Ltd.

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The Low-Resolution Structure Analysis of the Lens Protein γ-Crystallin

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The structure of a calf-lens protein, γ-crystallin fraction (II), has been determined at low resolution (5·5 Å) using single-crystal X-ray diffraction methods. Phases for the diffraction data were calculated by the method of multiple isomorphous replacement and the use of anomalous scattering; heavy-atom derivatives were obtained using ethyl mercury thiosalicylate, ethyl mercury chloride, potassium auricyanide and uranyl acetate. The electron density map shows that the molecule is organized as two globular domains each approximately 25 Å in radius. Preliminary crystallographic observations on two other calf γ-crystallins, fractions (III*b*) and (IV), are also reported.

Introduction

The cytoplasm of the fibrous cells of the vertebrate eye lens contains lens specific proteins, the crystallins. In mammals they are heterogeneous and are classified as α-, β- and γ-crystallins; the relative proportions reflect the region of the lens from which they were isolated (see review by Harding & Dilley, 1976). The γ-crystallins are a family of monomeric proteins of molecular weight 20 000 found mainly in the central core region or 'nucleus' of the lens. Amino acid sequence studies have shown that the different γ-crystallin fractions (II), (III*a*), (III*b*), and (IV) are closely related (Croft, 1972, 1973; Slingsby & Croft, 1978). Preliminary crystallographic investigations on fraction (II) have been reported by us (Carlisle, Lindley, Moss & Slingsby, 1977) and on fractions (II) and (III*b*) by Chirgadze, Nikonov, Garber & Reshetnikova (1977).

The tertiary structure of a structural protein should indicate those features of the polypeptide chain folding which enable the protein to have high stability. This is of particular importance to the γ-crystallins as these proteins are synthesized mainly during the embryonic and foetal stages of mammalian development and are retained in the centre of the eye lens with very little protein turnover for the lifespan of the animal. In long-lived species, *e.g.*, man, the central regions frequently become less transparent in old age with the development of senile nuclear cataract. In cataract the proteins of the human lens nucleus gradually denature, leading to polymerization, pigmentation and acquisition of unknown covalent crosslinks other than disulphide bonds (Dilley & Pirie, 1974). Recent chemical analyses clearly indicate the association of cataract with oxidation of protein sulphydryl to protein–protein disulphide bonds (Takemoto & Azari, 1976), this association being particularly striking in the lens-nuclear region (Truscott & Augusteyn, 1977).

One of the aims of the tertiary structure analysis of the bovine γ-crystallin is to further define the environment and reactivity of the six cysteine residues of fraction (II) and to compare these regions with the corresponding regions in the other γ-crystallin fractions. We present here a low-resolution (5·5 Å) three-dimensional structure analysis of γ-crystallin fraction (II) and preliminary investigations of γ-crystallin fractions (III*b*) and (IV).

Experimental

(*a*) *Extraction*

Isolation of calf γ-crystallin and its separation into fractions (I), (II), (III) and (IV) was achieved by methods previously described (Carlisle *et al.*, 1977). A new method of separation of γ-crystallin fraction (III) was devised. To a column of DEAE-cellulose (55 × 2