# Structure of Poly-L-Proline II

By V. Sasisekharan

*Department of Physics, University of Madras, Madras-25, India*

The structure of poly-L-proline is completely worked out by X-ray and optical diffraction methods. The polymer chains are arranged in a hexagonal cell with $a = 6.68$ and $c = 9.36$ Å. The space group is $P3_2$. The coordinates of the atoms were determined by accurate graphical methods. The correctness of the structure was also verified by intensity calculations. For this arrangement of the polymer chains, there are the least number of short contacts, the only one which occurs being a C–H $\cdots$ O hydrogen bond. From packing considerations also the structure arrived at is seen to be the best.

## 1. Introduction

This paper and the succeeding one on the structure of poly-L-hydroxyproline deal with the crystalline structure of certain forms of the two polypeptides which are related to the structure of the protein, collagen. Collagen is characterized by a large abundance of these imino-acid residues and, in particular, it is the only protein in which hydroxyproline occurs to a significant extent. A study of these two polypeptides is therefore likely to help in working out fully the structure of collagen.

Unfortunately, neither poly-L-proline nor poly-L-hydroxy proline has a unique structure in its solid state. Even the powder pattern of the precipitate shows appreciable changes according to the solvent used and the way in which the precipitate was obtained. Changes in optical activity with time in water and other solutions have already been reported for poly-L-proline (Kurtz *et al.*, 1956, 1958; Harrington & Sela, 1958; Blout & Fasman, 1958). A paper on the structural changes in these two polypeptides as revealed by X-ray, optical activity and other studies made by the author, will be published elsewhere. In this paper and the next one, the discussion is restricted to one modification of each of the two polypeptides, which are somewhat similar to each other. Both give X-ray patterns broadly resembling each other. Both also show a large negative rotation (specific rotation is $[\alpha]_D^{25} = -400°$ in water and $[\alpha]_D^{27} = -260°$ in acetic acid for poly-L-proline II and poly-L-hydroxyproline *A* respectively), similar to collagen and gelatin.

A triple helical structure for collagen is now generally accepted (Ramachandran & Kartha, 1955; Ramachandran, 1956; Rich & Crick, 1958) and it is reasonable to suppose that these two polypeptides will also have similar chains in the unit cell. In fact, at about the time Ramachandran & Kartha (1955) put forward their first model of collagen, Cowan & McGavin (1955) suggested a hexagonal structure for poly-L-proline II with a helical chain of three residues having a three fold screw axis symmetry per unit cell. The helix is a left-handed one (space group $P3_2$) with an axial repeat of 9.36 Å about the unique (fibre) axis. The sense of the helix is uniquely fixed from the absolute configuration of the groups around the $\alpha$-carbon atom as found in L-amino acids (Bijvoet, Peerdemann & van Bommel, 1951; Raman, 1958). It is found that a right-handed helix cannot be built with the dimensions required to fit the X-ray data.

Although Cowan & McGavin (1955) showed that a left-handed helix can be constructed with a $c$ axis repeat of 9.36 Å and made discussion on the two packing positions of the helices in the unit cell, they gave no definite values of the atomic parameters. No further work appears to have been reported about the arrangement of the chains in the unit cell or regarding the detailed check of calculated and observed intensities. This is done in the present paper.

We assume that the configuration of a single chain of the polymer is as mentioned above and we are interested in finding out how the chains are packed in the unit cell. The nitrogen atoms of the prolyl residues carry no hydrogen atoms to form N–H...O=C hydrogen bonds. Therefore, other stereochemical factors should play an important role in determining the molecular arrangement. It is actually found that there is an arrangement of the polymer chains in the unit cell with uniform density of packing which could at the same time account for the intensities of the X-ray reflections as well. For this arrangement, there is a short contact of the order of 3.0 Å between the $\gamma$ carbon of the prolyl residue of one chain and carbonyl oxygen of another chain. There is no other short contact of any sort. It appears that the structure of this polypeptide is stabilized by the formation of C–H ... O hydrogen bonds, one for each oxygen present.

## 2. Redetermination of the molecular structure of a single chain

Before finding the best orientation of the chains, the coordinates of the atoms in a single chain were recalculated fairly accurately by graphical methods. X-ray powder and oriented-film patterns were ob-

tained by the author for the sample of poly-L-proline II, kindly supplied by Prof. Katchalski. These results confirmed the space group and unit cell of poly-L-proline II already reported. The unit-cell dimensions are $a = 6 \cdot 68$ Å, which is slightly higher than the value previously reported, viz. $a = 6 \cdot 62$ Å, and the fibre axis repeat distance $c = 9 \cdot 36$ Å. The X-ray powder pattern is shown in Fig. 1 and the observed and calculated $d$ spacings of the reflections are given in Table 1. The observed density was $1 \cdot 32$ g.cm.$^{-3}$ which agreed well with the calculated value $1 \cdot 34$ g.cm.$^{-3}$.
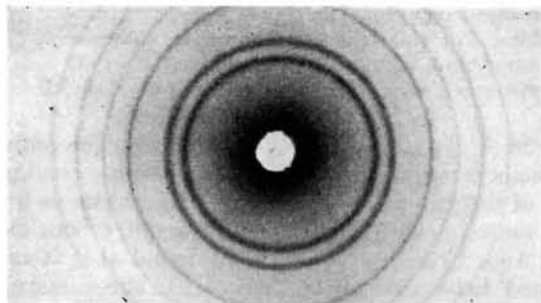


Fig. 1. X-ray powder pattern of poly-L-proline II taken with a cylindrical camera of radius $5 \cdot 73$ cm.

In deriving the coordinates of the backbone, the peptide dimensions as given by Corey & Pauling (1953) were strictly adhered to. The residue height is $3 \cdot 12$ Å. As there should be three residues per turn, the radius of the helix on which the $\alpha$-carbon atoms lie is $1 \cdot 25$ Å. This fixes the positions of the $\alpha$-carbon atoms. The orientation of the proline residue depends upon the orientation of the peptide with respect to the helical axis. The peptide should be rotated about the line $\alpha C_1 - \alpha C_2$, through nearly 90° from the vertical position (as for instance in the $\alpha$ helix) so that the bond angle $N_1 - \alpha C_1 - C_2'$ is within the range 105° to 115°.

If the peptide is rotated exactly through 90° from the vertical position, this bond angle is 110° 30'. For this position, the unbonded distance $N_1 O_2$ within the chain is $2 \cdot 80$ Å which is just the normal contact distance between N and O. If the bond angle is reduced, then the $N_1 O_2$ distance is also reduced. On the other hand, if the bond angle $N_1 - \alpha C_1 - C_2'$ is increased, the position of the $\beta$ carbon of the proline residue has to be moved considerably out of the plane containing the remaining four atoms of the ring, though the unbonded distance $N_1 O_2$ increases. Otherwise, there will be a short contact between $O_2$ and $\beta C_1$ within the chain. When the bond angle is 110° 30', the $\beta$ carbon has to be moved about $0 \cdot 40$ Å out of the plane such that the distance between $O_2$ and $\beta C_1$ within a chain is $3 \cdot 25$ Å. This seems to be reasonable, and for this position the coordinates of the atoms in the peptide were determined.

The proline residue could then be easily attached to the backbone. First a model based on the coordinates of the proline residue as given by Mathieson & Welsh (1952) was used. In this model the $\gamma$ carbon atom is very much above the plane containing the remaining four atoms in the ring. A chain based on this could not be accommodated in a unit cell of $a = 6 \cdot 68$ Å and $c = 9 \cdot 36$ Å, for there were severe short contacts between the various atoms of different chains. The strain is relieved if the height of the $\gamma$ carbon atom above the plane containing the remaining atoms is decreased. In fact, if a planar residue is attached to the backbone, the chains can be packed well in the unit cell. In the model shown in Figs. 4 and 5 the proline residue is almost planar except for the $\beta C$ which is about $0 \cdot 4$ Å above the plane. The $\gamma$ carbon atom is very slightly pushed below the plane.

Table 1. *Observed and calculated d spacings of the reflections of poly-L-proline II*

| No. | $d_o$ (Å) | $d_c$ (Å) | $hkl$ |
|---|---|---|---|
| 1 | 5·76 | 5·77 | 100 |
| 2 | 4·90 | 4·90 | 101 |
| 3 | 3·65 | 3·64 | 102 |
| 4 | 3·35 | 3·34 | 110 |
| 5 | 3·13 | 3·12 | 003 |
|   |   | 3·14 | 111 |
| 6 | 2·88 | 2·89 | 200 |
| 7 | 2·75 | 2·74 | 103 |
| 8 | 2·71 | 2·72 | 112 |
| 9 | 2·45 | 2·46 | 202 |
| 10 | 2·28 | 2·28 | 113 |
| 11 | 2·15 | 2·17 | 104 |
|   |   | 2·13 | 211 |
| 12 | 1·93 | 1·93 | 300 |
|   |   | 1·92 | 114 |
| 13 | 1·79 | 1·78 | 302 |
|   |   | 1·79 | 213 |
| 14 | 1·58 | 1·57 | 222 |
|   |   | 1·58 | 311 |
| 15 | 1·50 | 1·51 | 312 |

Table 2. *Cylindrical coordinates of the atoms in a single residue of poly-L-proline II, obtained by the author and by Cowan & McGavin*

| Atom | Author | | | Cowan & McGavin | | |
|---|---|---|---|---|---|---|
|  | $r$ (Å) | $\theta$ (°) | $z$ (Å) | $r$ (Å) | $\theta$ (°) | $z$ (Å) |
| $\alpha C_3$ | 1·25 | 0° 00' | 0·00 | 1·30 | 0° 00' | 0·00 |
| $C_1$ | 0·27 | 20° 30' | 1·16 | 0·32 | 12° 30' | 1·14 |
| $N_1$ | 1·01 | −75° 30' | 1·94 | 1·01 | −76° 00' | 1·95 |
| $O_1$ | 1·18 | 112° 30' | 1·33 | 1·14 | 109° 30' | 1·28 |
| $\alpha C_1$ | 1·25 | −120° 00' | 3·12 | 1·30 | −120° 00' | 3·12 |
| $\beta C_1$ | 2·65 | −105° 30' | 3·44 | 2·68 | −103° 30' | 3·44 |
| $\gamma C_1$ | 3·19 | −81° 00' | 2·78 | 3·17 | −77° 00' | 2·86 |
| $\delta C_1$ | 2·47 | −64° 30' | 1·70 | 2·44 | −63° 00' | 1·75 |

The positions of the atoms as determined by the author are given in polar coordinates in Table 2 along with the coordinates derived by Cowan & McGavin (1955). The discrepancy in the coordinates of the atoms in the peptide plane may be attributed to the fact that the earlier authors might not have strictly adhered to the Corey–Pauling dimensions for the peptide. Or perhaps the coordinates were obtained from model building and not from accurate graphical construc-

tions. In any case, the structure of the individual chains is very similar in both.

## 3. Determination of the coordinates of the atoms in the unit cell

Since the nitrogen atoms carry, as already mentioned, no hydrogen atoms for N–H $\cdots$ O = C hydrogen bonds to be formed, the molecular arrangement is chiefly determined by other stereochemical factors. To study these, the poly-L-proline chains were rotated about the fibre axis in the unit cell and the short contacts between the neighbouring chains were all listed. The stereochemically best structure could then be picked out.

### (a) Listing of short contacts

To start with, the chains were kept with an $\alpha$ carbon atom lying on the $a$ axis. This may be denoted by an angle $\psi = 0°$ as shown in Fig. 2. From this position, the helices were then turned through intervals of $5°$ in the clockwise direction, the short contacts being listed for each value of $\psi$, namely $0°$, $5°$, $10°$, $\ldots$, $55°$. In the present study, these distances were determined by projecting the molecules on to the $ab$ plane and plotting the atoms on a scale of 2 in. equal to 1 Å. Measuring off the projected distance and knowing the difference in the $z$ coordinates, the distance between two atoms can be calculated. It is enough to rotate the chain through $60°$ in all to take into account all possibilities. In Table 3 are listed the various short contacts and it will be seen that no position is completely devoid of short contacts. At the beginning (for small $\psi$) there are short contacts between the carbon atoms with a low value of 3·20 Å between the $\delta C_1$ of the first chain and $\gamma C_3$ of the next chain. There are also short contacts between the oxygen and the carbon atoms. The oxygen $O_3$ of the first chain is away from the $\delta C_3$ and $\gamma C_3$ by 3·07 Å and 3·14 Å respectively. As the helix is rotated the distances
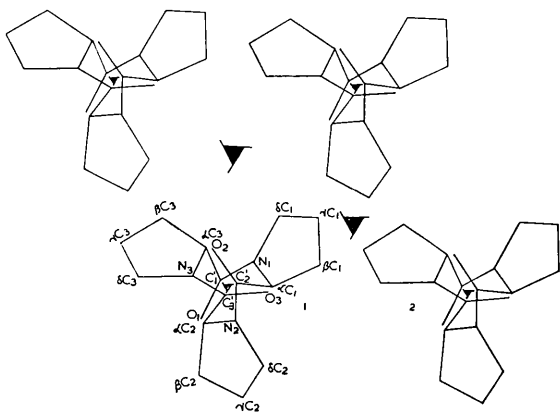


Fig. 2. Projection of the molecular chain on the $a$, $b$ plane. The position indicated is $\psi = 0°$ from which the chain is rotated through intervals of $5°$.

Table 3. *List of short contacts as the helix is rotated from* $\psi = 0$ *to* $\psi = 55°$

C–C contacts greater than 3·6 Å are omitted

| $\psi°$ | Atom in first chain | Atom in second chain | Distance (Å) |
|---|---|---|---|
| 0 | $\gamma C_1$ | $\delta C_3$ | 3·40 |
| | $O_3$ | $\gamma C_3$ | 3·14 |
| | $\delta C_1$ | $\gamma C_3$ | 3·20 |
| | $O_3$ | $\delta C_3$ | 3·07 |
| | $\gamma C_1$ | $\beta C_3$ | 3·40 |
| 5 | $\gamma C_1$ | $\gamma C_3$ | 3·37 |
| | $\gamma C_1$ | $\beta C_3$ | 3·40 |
| | $\delta C_1$ | $\gamma C_3$ | 3·10 |
| | $O_3$ | $\gamma C_3$ | 3·40 |
| | $O_3$ | $\delta C_3$ | 3·15 |
| 10 | $\gamma C_1$ | $\gamma C_3$ | 3·32 |
| | $\delta C_1$ | $\gamma C_3$ | 3·01 |
| | $O_3$ | $\delta C_3$ | 3·31 |
| 15 | $\gamma C_1$ | $\gamma C_3$ | 3·38 |
| | $\delta C_1$ | $\gamma C_3$ | 3·00 |
| 20 | $\delta C_1$ | $\gamma C_3$ | 3·10 |
| | $\gamma C_1$ | $\gamma C_3$ | 3·55 |
| 25 | $\delta C_1$ | $\gamma C_3$ | 3·25 |
| | $\gamma C_1$ | $O_1$ | 3·20 |
| 30 | $\delta C_1$ | $\gamma C_3$ | 3·55 |
| | $\gamma C_1$ | $O_1$ | 3·00 |
| 35 | $\gamma C_1$ | $O_1$ | 2·85 |
| 40 | $\gamma C_1$ | $O_1$ | 2·76 |
| | $\delta C_1$ | $O_1$ | 3·28 |
| 45 | $\gamma C_1$ | $O_1$ | 2·75 |
| | $\delta C_1$ | $O_1$ | 3·14 |
| 50 | $\gamma C_1$ | $O_1$ | 2·83 |
| | $\delta C_1$ | $O_1$ | 3·06 |
| 55 | $\delta C_1$ | $\gamma C_2$ | 3·43 |
| | $\gamma C_1$ | $O_1$ | 2·98 |
| | $\delta C_1$ | $O_1$ | 3·03 |

between the oxygen atoms and the carbon atoms increase whereas the distances between the carbon atoms decrease. In fact, at $\psi = 15°$ the distance between $\delta C_1$ of one chain and $\gamma C_3$ of another chain is as low as 3·00 Å. And there is no short contact between the oxygen atoms and the carbon atoms. As the helix is further rotated, the distance between the carbon atoms increases whereas the distance between $\gamma C_1$ of one chain and $O_1$ of the other chain decreases. At $\psi = 35°$, this distance is 2·85 Å and there is no other short contact. This value decreases and reaches a minimum at $\psi = 45°$, when the value is 2·75 Å. From this stage onwards, $\delta C_1$ of one chain is also close to $O_1$ of the other. The carbon atoms also come closer to each other. Thus we see that there are either short contacts between oxygen and carbon atoms or between carbon atoms themselves or both are present. It looked as though it may not be possible to arrive at the best structure from stereochemical considera-

Table 4. *List of intensities of the reflections in the hk0 and h0l projections as obtained from the optical diffraction patterns for $\psi = 0°$ to $55°$ positions*

The intensities of the hk0 and hk̄0 reflections, and h0l and h0l̄ reflections were respectively added.
The last column gives the observed intensities of the reflections in the X-ray powder pattern

| Reflection | 0 | 5 | 10 | 15 | 20 | 25 | 30 | 35 | 40 | 45 | 50 | 55 | X-ray |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 100 | s | s | s | s | s | s | s | s | s | s | s | s | s |
| 200 | m | m | w | w | vw | vw | w | w | w | m | m | m | vw |
| 300 | — | — | — | w | w | m | w | w | vw | — | — | — | w |
| 400 | — | m | m | w | — | w | w | — | w | m | w | — | — |
| 110 | w | w | w | w | m | m | w | w | w | w | w | w | vw |
| 120 | m | w | w | w | w | w | w | m | m | m | w | w | — |
| 130 | w | w | m | s | m | w | — | w | m | m | w | w | — |
| 220 | s | m | w | — | — | — | — | w | w | — | w | m | — |
| 230 | w | m | s | m | s | m | w | w | w | — | w | — | w |
| 003 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| 101 | s | m | m | s | s | s | s | s | s | s | s | s | s |
| 102 | m | m | m | s | s | s | s | s | s | s | s | s | s |
| 103 | m | m | m | m | s | s | s | s | s | s | m | m | m |
| 104 | w | w | m | w | w | w | w | w | vw | vw | vw | vw | vw |
| 201 | — | — | — | — | — | — | — | — | vw | vw | vw | — | — |
| 202 | vs | s | m | w | w | — | vw | w | m | s | vs | vs | vw |
| 203 | — | w | m | m | s | m | w | vw | vw | vw | — | — | — |
| 204 | m | m | m | m | w | w | — | — | — | vw | w | m | — |
| 301 | vw | — | — | — | w | — | — | vw | vw | vw | w | vw | — |
| 302 | w | w | vw | w | vw | vw | vw | vw | w | m | w | — | vw |
| 303 | w | — | vw | vw | vw | vw | w | w | vw | w | m | m | — |
| 304 | s | s | m | m | m | vw | vw | vw | vw | vw | w | m | — |
| 401 | w | w | vw | vw | vw | vw | vw | vw | vw | vw | vw | w | — |
| 402 | w | w | vw | w | w | w | w | w | vw | vw | vw | vw | — |
| 403 | w | — | vw | — | vw | w | m | w | vw | w | w | w | — |
| 404 | w | — | vw | — | vw | — | w | m | m | m | m | w | — |

*vs*—very strong; *s*—strong; *m*—medium; *w*—weak; *vw*—very weak.

tions alone. Therefore, intensity calculations for these arrangements were optically made and compared with the X-ray data.

### (b) Optical transforms and their comparison with X-ray data

The positions of the atoms in the twelve cases mentioned above were plotted on the *ab* and *ac* planes. Punched masks were obtained corresponding to these plottings and the diffractometer constructed in this laboratory (Aravindakshan, 1957) was used to obtain the optical transforms of the punched masks. The strongest reflections and other relevant facts obtained from the optical diffraction patterns of both hk0 and h0l projections are given in Table 4.

Among all the reflections observed in the X-ray powder pattern (Fig. 1) the 100 and 101 reflections are the strongest in intensity while the 102 reflection is moderately strong. The reflections 112 and 103 combine to form a broad ring and are of medium intensity. Among the hk0 reflections 100 is the strongest. Reflections 300, 200 and 110 are weak and are of the same order.

While comparing the X-ray pattern with the optical diffraction data, one should bear in mind the fact that the optical diffraction experiments directly give the square of the structure amplitude, whereas the intensities of the reflections in the powder pattern are due to the product of the square of the structure

amplitudes with Lorentz, polarization and temperature factors.

On comparing the hk0 reflections, agreement between the X-ray data and the optical patterns is seen to be good only for a range between $\psi = 25°$ and $\psi = 35°$. At $\psi = 30°$ the agreement is best. Comparing the h0l reflections, it is observed that there is agree-

Table 5

(a) Coordinates of the atoms in a single residue of poly-L-proline II molecule

| Atom | $x$ | $y$ | $z$ |
|---|---|---|---|
| $\alpha C_1$ | 1·108 | −0·108 | 0·333 |
| $C_1'$ | 0·008 | 0·044 | 0·120 |
| $N_1$ | 0·168 | 0·044 | 0·207 |
| $O_1$ | 0·202 | −0·078 | 0·142 |
| $\beta C_1$ | 0·321 | −0·122 | 0·368 |
| $\gamma C_1$ | 0·515 | 0·086 | 0·297 |
| $\delta C_1$ | 0·426 | 0·183 | 0·182 |

(b) List of bond lengths and bond angles in the proline residue of the chain

| Bond | Length | Atoms | Angle |
|---|---|---|---|
| $\alpha C_1 - \beta C_1$ | 1·52 Å | $\alpha C_1 - N_1 - \delta C_1$ | 115° 00′ |
| $\alpha C_1 - N_1$ | 1·47 | $N_1 - \delta C_1 - \gamma C_1$ | 103 00 |
| $\beta C_1 - \gamma C_1$ | 1·51 | $\delta C_1 - \gamma C_1 - \beta C_1$ | 111 00 |
| $\gamma C_1 - \delta C_1$ | 1·50 | $\gamma C_1 - \beta C_1 - \alpha C_1$ | 107 00 |
| $\delta C_1 - N_1$ | 1·51 | $C_2' - \alpha C_1 - \beta C_1$ | 112 00 |
| | | $C_2' - \alpha C_1 - N_1$ | 110 30 |
| | | $C_1' - N_1 - \delta C_1$ | 125 00 |

The bond angles and bond distances in the backbone of the peptide are as given by Corey & Pauling (1953).

hk0 projection

$\psi \rightarrow$ 25° 30° 35° 40°



h0l projection

$\psi \rightarrow$ 25° 30° 35° 40°
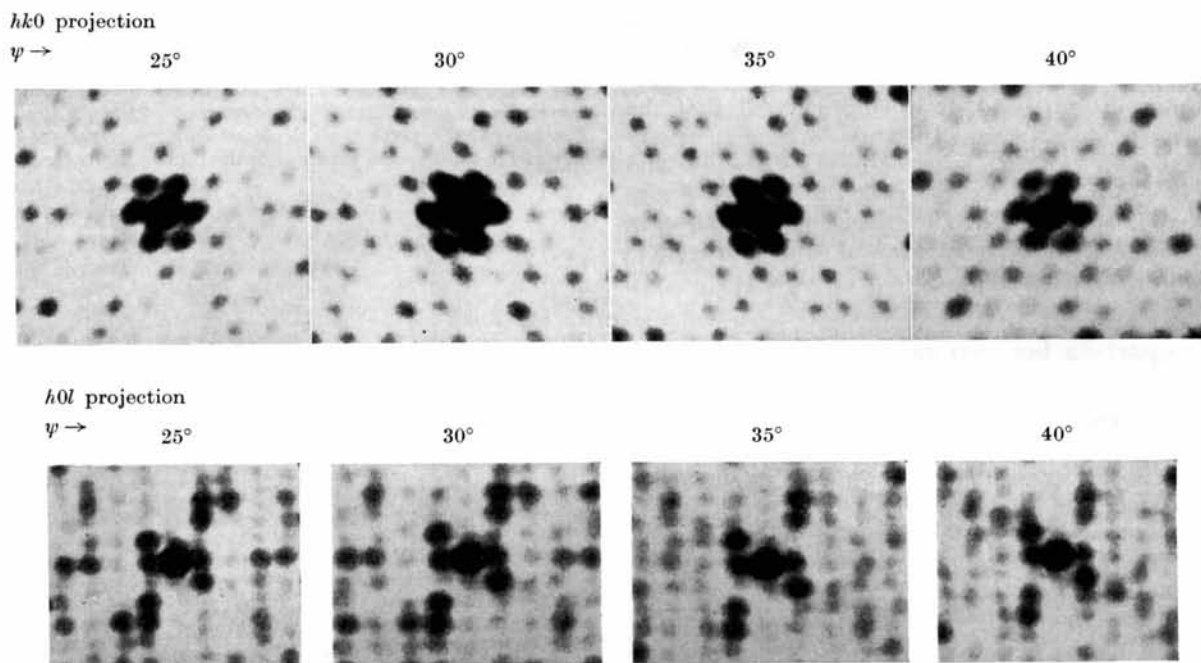


Fig. 3. Optical diffraction patterns corresponding to the positions of $\psi = 25°$, 30°, 35°, and 40°
for both hk0 and h0l projections respectively.

ment between the X-ray data and the optical diffraction data for a range from $\psi = 25°$ to $\psi = 40°$. The best agreement seems to be at $\psi = 35°$. The optical transforms corresponding to $\psi = 25°$, 30°, 35°, and 40° for the two projections are shown in Fig. 3.

It is clear that the arrangement of the molecules in the unit cell should be between the $\psi = 30°$ and 35° positions. Referring to Table 3, it is observed that these positions correspond also to the minimum number of short contacts. In fact, there is only one such short distance between the $\gamma C_1$ of one chain and $O_1$ of another chain in both cases (leaving aside the $\delta C_1 - \gamma C_3$ distance of 3·55 Å for $\psi = 30°$, which is close enough to 3·6 Å, the accepted value). Of the two,

$\psi = 30°$ position is preferable as the distance from $\gamma C_1$ of one chain to $O_1$ of another chain is 3·0 Å longer than for $\psi = 35°$. Thus there might be a hydrogen bond between the $CH_2$ group and an oxygen atom, since it is appreciably shorter than the unbonded C–O distance. Assuming that the hydrogen bond exists, the bond angles were calculated to be as follows: At $\psi = 30°$ the angle $\delta C_1 - \gamma C_1 - O_1'$ is 106° and $\beta C_1 - \gamma C_1 - O_1'$ is 108° ($O_1'$ indicates $O_1$ of another chain). At $\psi = 35°$ the angle $\delta C_1 - \gamma C_1 - O_1'$ is 101° and $\beta C_1 - \gamma C_1 - O_1'$ is 117°. At $\psi = 40°$ $\delta C_1 - \gamma C_1 - O_1'$ is 95° and $\beta C_1 - \gamma C_1 - O_1'$ is 130°. Thus it is observed that the bond angle is also favourable for such a hydrogen bond to
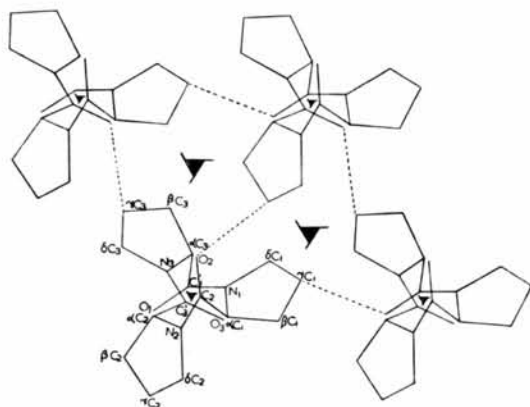


Fig. 4. Structure of poly-L-proline II projected on the a, b plane. C–H · · · O hydrogen bonds are shown by dashed lines.
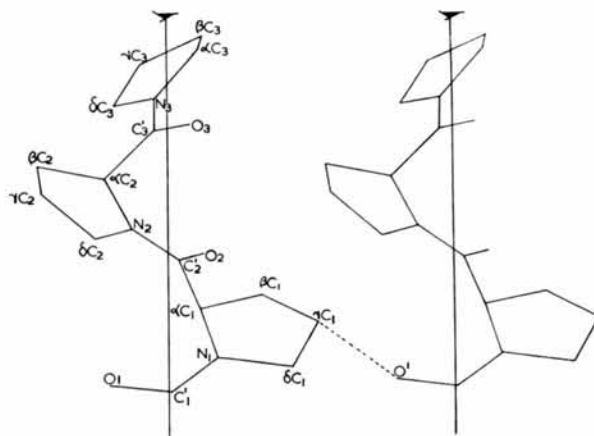


Fig. 5. Structure of poly-L-proline II projected on the a, c plane. The C–H · · · O hydrogen bond is shown by a dashed line.

be formed, and the various angles at the carbon atom are close to the tetrahedral angle at $\psi=30°$. If we assume that the bond angles should be close to $110°$ for the hydrogen bond, it will be deduced that the structure of poly-L-proline II should correspond closely to the $\psi=30°$ position, where $\delta C_1-\gamma C_1-O_1' = 106°$ and $\beta C_1-\gamma C_1-O_1' = 108°$. In that case the bond distance C–H $\cdots$ O will be $3\cdot0$ Å. The arrangement is shown in Figs. 4 and 5. The coordinates of the atoms thus arrived at are given in Table 5.

## 4. Comparison between calculated and observed intensities

Using the coordinates of the atoms for the $\psi=30°$ position given in Table 5, the above conclusions regarding the X-ray intensities were verified by intensity calculations. The intensities of all the possible reflections which can occur with $h^2+k^2<18$ and with $l$ index up to 4 were calculated.

The scattering factors for C, N, and O atoms were taken from the reported values (Viervoll & Øgrim, 1949). Because of the hexagonal symmetry, the intensities of the $h0l$ series are identical with the intensities of the corresponding reflections of the $0kl$ series. The calculated values of the intensities of the reflections are tabulated in Table 6 along with the observed intensities. The calculated values have been multiplied by Lorentz and polarization factors and also by an isotropic temperature factor with $B=4$ Å², the value usually found in organic crystals. The agreement between the calculated and observed intensities is seen to be good. In no case has a strong reflection predicted by calculation not been observed. In the last column of Table 6 are also given the calculated squares of the structure factors for comparison with the optical diffraction data for the $\psi=30°$ position (Fig. 3).

## 5. Conclusion

The structure of poly-L-proline II has thus been completely worked out. The fractional coordinates of the atoms in one residue with the bond lengths and bond angles corresponding to the structure at $\psi=30°$ are given in Table 5. The bond angles and bond distances are within normal limits. No further attempt to refine the coordinates has been made in the absence of accurate intensity data.

A C–H $\cdots$ O bond of length $3\cdot0$ Å is found to occur in this structure. Such a bond is in fact to be expected, for there is no other attractive forces to connect the polymer chains to one another and thus stabilize the structure. Otherwise the strong electronegative carbonyl oxygen is left unbonded. Such C–H $\cdots$ O bonds have, in fact, been previously reported (Hunter, 1946; Donohue & Trueblood, 1952).

From packing considerations also, the arrangement shown seems to be the best. In no place is there a region of low density. The protuberances of one chain

Table 6. *Calculated and observed intensities of the reflections of poly-*L-*proline II*

The calculated intensities are multiplied by Lorentz and polarisation factors and a temperature factor with $B=4$ Å². The last column gives the calculated squares of the structure factors for comparison with the optical diffractions for $\psi=30°$

| hkl | $I_o$ | $I_c \times 10^{-2}$ | $|F|^2_c$ |
|---|---|---|---|
| 100 | s | 1668 | 1632 |
| 101 | s | 928 | 1310 |
| 102 | ms | 422 | 1192 |
| 110 | vw | 52 | 289 |
| 003 ⎫ | m | ⎰ 24 | 99 |
| 111 ⎭ | | ⎱ 89 | 365 |
| 200 | vw | 42 | 214 |
| 201 | — | 32 | 185 |
| 103 ⎫ | broad | ⎰ 183 | 1071 |
| 112 ⎭ | medium | ⎱ 206 | 1241 |
| 202 | vw | 16 | 125 |
| 113 | vw | 38 | 384 |
| 210 | — | 22 | 250 |
| 104 ⎫ | w | ⎰ 24 | 280 |
| 211 ⎭ | | ⎱ 71 | 889 |
| 203 | — | 14 | 175 |
| 212 | — | 39 | 623 |
| 300 ⎫ | w | ⎰ 16 | 287 |
| 114 ⎭ | | ⎱ 39 | 698 |
| 301 | — | 9 | 168 |
| 204 | — | 15 | 318 |
| 213 ⎫ | vw | ⎰ 18 | 412 |
| 302 ⎭ | | ⎱ 6 | 143 |
| 220 | — | 2 | 76 |
| 221 | — | 7 | 208 |
| 303 | — | 11 | 345 |
| 310 | — | 2 | 57 |
| 214 | — | 7 | 239 |
| 311 ⎫ | vw | ⎰ 8 | 305 |
| 222 ⎭ | | ⎱ 11 | 419 |
| 312 | vw | 14 | 601 |
| 304 | — | 2 | 80 |
| 223 | — | 9 | 46 |
| 400 | — | 4 | 207 |
| 401 | — | 1 | 71 |
| 313 | — | 5 | 276 |
| 402 | — | 2 | 119 |
| 224 | — | 8 | 585 |
| 320 | — | 3 | 223 |
| 314 | — | 8 | 670 |
| 321 | — | 3 | 244 |
| 403 | — | 2 | 135 |
| 322 | — | 5 | 424 |
| 323 | — | 2 | 258 |
| 404 | — | 1 | 114 |
| 324 | — | 3 | 454 |

*s*—strong; *ms*—moderately strong; *m*—medium; *w*—weak; *vw*—very weak

nicely go into the vacant places left by another and so on. An even more uniform density of packing can be obtained if the helix is rotated through 2 or 3° more from the position indicated. However, such a refinement cannot be made with certainty until more exact X-ray data are available.

Thus the proposed structure, in short, has the following merits:

(a) Among all the possible orientations of individual chains, it agrees best with X-ray intensity data.

(b) At the same time, there are the least number of

short contacts, the only one which occurs being ac-
tually a C–H · · · O hydrogen bond.

(c) There is a uniform density of packing, the
'knobs' of one chain going into the 'holes' between the
other chains.

The author is indebted to Prof. G. N. Ramachan-
dran and wishes to express his sincere gratitude to
him for the keen interest, he took in this study. The
author is also grateful to Prof. E. Katchalski of the
Weismann Institute, Israel, for the sample of the
polymer which enabled him to carry out this study.
His thanks are due to the Government of India for
the award of a fellowship during the tenure of which
the work was carried out.

### References

ARAVINDAKSHAN, C. (1957). *J. Sci. Instrum.* **34**, 250.

BIJVOET, J. M., PEERDEMAN, A. F. & BOMMEL, A. J. VAN
(1951). *Nature, Lond.* **168**, 271.

BLOUT, E. R. & FASMAN, G. D. (1958). *Recent Advances in
Gelatin and Glue Research*, p. 123. London: Pergamon
Press.

COREY, R. B. & PAULING, L. (1953). *Proc. Roy. Soc.* B,
**141**, 10.

COWAN, P. M. & MCGAVIN, S. (1955). *Nature, Lond.* **176**,
501.

DONOHUE, J. & TRUEBLOOD, K. N. (1952). *Acta Cryst.*
**5**, 419.

HARRINGTON, W. F. & SELA, M. (1958). *Biochim. Bio-
phys. Acta*, **27**, 24.

HUNTER, L. (1946). *Rep. Progr. Chem.* **43**, 153.

KURTZ, J., BERGER, A. & KATCHALSKI, E. (1956). *Nature,
Lond.* **178**, 1066.

KURTZ, J., BERGER, A. & KATCHALSKI, E. (1958).
*Recent Advances in Gelatin and Glue Research*, p. 131.
London: Pergamon Press.

MATHIESON, A. McL. & WELSH, H. K. (1952). *Acta Cryst.*
**5**, 599.

RAMACHANDRAN, G. N. & KARTHA, G. (1955). *Proc. Ind.
Acad. Sci.* A, **42**, 215.

RAMACHANDRAN, G. N. (1956). *Nature, Lond.* **177**, 710.

RAMAN, S. (1958). Ph.D. Thesis (Madras).

RICH, A. & CRICK, F. H. C. (1958). *Recent Advances in
Gelatin and Glue Research*, p. 20. London: Pergamon
Press.

VIERVOLL, H. & Øgrim, O. (1949). *Acta Cryst.* **2**, 277.

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# Structure of Poly-L–Hydroxyproline *A*

BY V. SASISEKHARAN

*Department of Physics, University of Madras, Madras, 25, India*

The structure of poly-L-hydroxyproline *A* is worked out from X-ray powder and oriented film
patterns. The polymer chains are packed in a hexagonal lattice with $a = 12\cdot3$ and $c = 9\cdot15$ Å. The
space group is $P3_2$. The arrangement of the chains is shown to be very similar to that of poly-L-
proline II. It is also shown that the fundamental unit of the structure requires three helices as in
collagen. The correctness of the structure was also verified by intensity calculations. The agreement
between the calculated and observed intensities is good. No further refinement is carried out in the
absence of accurate intensity data.

## 1. Introduction

From the structural view point, poly-L-proline and
poly-L-hydroxyproline are of special interest. Although
proline is a normal component of many proteins,
hydroxyproline is found characteristically in colla-
geneous tissue proteins. These polymers as a conse-
quence of being imino acids, can only contribute to
hydrogen bonded structures through their $C = O$
groups, since the nitrogen of the prolyl residues carry
no hydrogen atoms. Also there is no free rotation
between the imino group and the $\alpha$ carbon atom and
this restricts the configurational possibilities. In fact,
the restricted rotation of the N–C$\alpha$ bond undoubtedly
determines the way in which the proteins, collagen
and gelatin which contain high proportions of these

imino acids, may form folded structures in the solid
state or in solution. The detailed investigation of
poly-L-proline II, reported in the previous paper,
has shown that this polymer resembles collagen and
gelatin in its X-ray pattern and high optical rotation.
As the polar properties of the hydroxyl group of the
hydroxyproline residue are supposed to play an im-
portant role in determining the behaviour of collagen
and gelatin, an X-ray investigation of the structure
of poly-L-hydroxyproline is of great interest.

Poly-L-hydroxyproline of high molecular weight has
been synthesised recently by Katchalski *et al.* (1956).
A sample of the polymer was kindly made available
by Prof. Katchalski. A detailed study of the X-ray
pattern and optical activity of this material made by