

## The Crystal Structure of DL-Proline Hydrochloride

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The crystal structure of DL-proline hydrochloride ( $C_5H_9NO_2 \cdot HCl$ ) has been determined by X-ray diffraction methods. It crystallizes in the monoclinic space group  $P2_1/a$ ,  $Z=4$ , with unit-cell dimensions  $a=12.49$ ,  $b=6.71$ ,  $c=8.70$  Å, and  $\beta=93^\circ 25'$ . Refinement was carried out by the full-matrix least-squares methods including anisotropic thermal parameters. Both the imino and carboxyl groups in the molecule are found to be protonated. The pyrrolidine ring is puckered at the  $\alpha$  carbon atom which deviates about 0.5 Å from the best plane formed by the four remaining atoms. The molecules are held together by a system of hydrogen bonds, with  $O \cdots Cl$  of 2.96,  $N \cdots Cl$  of 3.14 and 3.18 Å. The bond lengths and angles are compared with various related compounds.

### Introduction

Proline is a very important amino acid because of its unique conformation which may affect the structure of proteins, in particular collagen. However, unit-cell and space group examinations have so far been reported only for L-proline (Wright & Cole, 1949) and L-proline monohydrate (Sasisekharan, 1959). The crystal structure of the former has recently been reported by Kayushina & Vainshtein (1965). We have undertaken a three-dimensional crystal structure analysis of DL-proline hydrochloride in order to compare the mode of puckering of the pyrrolidine ring with other crystals containing proline or proline residues.

### Experimental

Crystals of DL-proline hydrochloride were obtained by dissolving a powder of DL-proline in an excess of con-

centrated hydrochloric acid and keeping the resulting solutions in a  $CaCl_2$  desiccator. The salt of the racemic mixture was gradually crystallized from the solution after a few days, while that of L-proline prepared in the same way remained in the state of viscous solution even after one month. The crystals of DL-proline hydrochloride were colourless plates with well developed {100} faces, with the longer edge parallel to  $b$  and the shorter edge parallel to  $c$ . The clear cleavage surface was (001). The crystals were cut into rods of diameter less than 0.2 mm and were sealed in Lindemann glass tubes.

Unit-cell dimensions were determined from precession photographs. The crystal system is monoclinic, with lattice constants,  $a=12.49 \pm 0.02$ ,  $b=6.71 \pm 0.01$ ,  $c=8.70 \pm 0.02$  Å,  $\beta=93^\circ 25' \pm 10'$ . The density measured by flotation in a mixed solution of carbon tetrachloride and toluene was  $1.373$  g.cm<sup>-3</sup>; the value assuming four units of  $C_5H_9NO_2 \cdot HCl$  per cell is  $1.384$  g.cm<sup>-3</sup>. The system-

Table 1. Fractional coordinates of the atoms and their standard deviations

	$x$	$10^4\sigma(x)$	$y$	$10^4\sigma(y)$	$z$	$10^4\sigma(z)$
Cl	0.1340	2.0	1.0508	3.9	0.1911	3.0
C(1)	0.4398	7.6	1.0053	13.6	0.1717	11.6
C(2)	0.4142	6.8	0.7895	14.3	0.2079	10.4
C(3)	0.4041	8.7	0.7514	19.5	0.3813	10.7
C(4)	0.3371	11.5	0.5647	25.1	0.3762	13.0
C(5)	0.2756	8.3	0.5512	19.7	0.2226	12.2
N	0.3121	5.3	0.7266	11.1	0.1291	8.4
O(1)	0.3894	5.4	1.1012	10.3	0.0695	7.9
O(2)	0.5188	4.8	1.0786	11.0	0.2627	7.1

Table 2. Final anisotropic temperature factors of the atoms ( $\times 10^3$ )

The temperature factor term is of the form

$$\exp [-(h^2\beta_{11} + k^2\beta_{22} + l^2\beta_{33} + 2hk\beta_{12} + 2kl\beta_{23} + 2hl\beta_{13})].$$

	$\beta_{11}$	$\beta_{22}$	$\beta_{33}$	$\beta_{12}$	$\beta_{13}$	$\beta_{23}$
Cl	3.97	9.03	8.72	1.88	1.46	1.13
C(1)	3.42	10.69	9.65	1.57	2.85	-0.67
C(2)	2.76	9.85	7.34	-0.55	0.63	1.16
C(3)	7.34	27.88	3.24	-7.06	-1.29	3.49
C(4)	15.22	54.59	8.95	-22.76	-2.89	11.55
C(5)	6.36	18.61	13.93	-3.75	-0.28	9.50
N	2.28	8.75	7.03	-1.25	-0.23	1.44
O(1)	6.01	15.61	9.96	-1.48	0.26	3.95
O(2)	4.10	16.93	10.09	-3.29	-1.15	-0.44

Table 3. Observed and calculated structure factors

M	K	L	F(OBS)	F(CAL)	M	K	L	F(OBS)	F(CAL)	M	K	L	F(OBS)	F(CAL)	M	K	L	F(OBS)	F(CAL)					
0	0	1	51.53	55.85	0	1	3	27.16	17.64	7	1	0	27.54	23.37	2	-3	21.28	-19.70	10	7	-4	6.44	-2.71	
0	0	2	83.99	-97.00	0	1	4	18.87	-14.77	7	1	1	34.20	30.99	2	-2	29.81	24.12	10	7	-3	6.58	5.81	
0	0	3	60.80	-59.53	0	1	5	23.89	-20.13	7	1	2	7.91	-3.68	2	-1	64.77	62.18	10	7	-2	10.41	15.63	
0	0	4	15.01	-11.76	0	1	6	12.38	-13.24	7	1	3	24.10	-22.08	2	0	3.79	-5.72	10	7	-1	9.26	9.26	
0	0	5	45.80	43.71	0	1	9	10.43	6.52	7	1	4	14.47	-6.35	2	1	34.67	-35.14	10	7	0	21.95	-21.97	
0	0	6	6.32	0.70	0	1	10	5.21	4.57	7	1	5	25.12	25.51	2	2	23.55	-22.42	10	7	1	15.50	-12.53	
0	0	7	22.21	-22.64	0	1	-1	11.13	-10.30	7	1	6	21.60	21.70	2	3	17.11	6.73	10	7	2	6.34	6.03	
0	0	8	9.38	-7.53	0	1	2	6.39	-4.95	7	1	7	8.81	-7.14	2	4	27.42	25.52	10	7	3	21.45	-15.18	
0	0	9	10.24	-11.14	0	1	3	11.52	11.57	7	1	8	13.87	-11.77	2	5	4.24	-5.84	10	7	4	16.10	15.39	
0	0	10	7.45	10.04	0	1	4	27.35	27.15	7	1	9	4.05	-4.96	2	6	-1.56	-11.30	10	7	5	8.90	-8.95	
0	0	11	5.87	0.95	0	1	5	22.58	20.74	7	1	10	3.47	-2.28	2	7	9.48	8.31	10	7	6	10.80	-8.81	
0	0	12	4.78	-4.99	0	1	6	31.31	-29.59	7	1	11	4.10	-1.60	2	8	6.29	35.18	10	7	7	5.50	-4.24	
2	0	-9	22.27	-21.61	1	1	-3	19.09	-18.26	8	-1	-5	2.00	4.29	2	10	-2.25	-2.25	11	7	-6	4.16	-4.15	
2	0	-8	5.88	-2.00	1	1	-2	41.63	41.85	8	-1	-4	8.68	-7.27	2	11	12.28	0.84	11	7	-5	10.39	8.74	
2	0	-7	19.08	24.37	1	1	-1	3.98	18.39	8	-1	-3	2.93	2.18	2	12	-1.20	9.49	11	7	-4	12.60	-12.05	
2	0	-6	4.40	-4.61	1	1	0	12.51	-11.73	8	-1	0	15.88	12.48	2	13	14.82	14.82	11	7	-3	9.48	-15.98	
2	0	-5	15.31	-15.68	1	1	1	29.07	-36.30	8	-1	1	17.30	16.10	2	14	4.72	-1.83	11	7	-2	4.38	0.28	
2	0	-4	58.09	48.63	1	1	2	46.99	-51.02	8	-1	2	6.91	1.24	2	15	11.16	25.87	11	7	-1	4.87	1.84	
2	0	-3	2.51	-31.00	1	1	3	16.46	-21.70	8	-1	3	28.27	-27.01	2	16	12.46	-9.70	11	7	0	15.20	-15.20	
2	0	-2	33.23	-24.33	1	1	4	4.25	4.55	8	-1	4	20.03	-17.99	2	17	37.80	35.51	11	7	1	16.37	-17.50	
2	0	-1	10.25	12.15	1	1	5	27.50	25.50	8	-1	5	10.71	9.54	2	18	45.68	-41.67	11	7	2	15.20	15.24	
2	0	0	21.92	14.76	1	1	6	8.60	6.10	8	1	7	4.54	-2.50	2	19	12.70	-10.65	11	7	3	14.53	13.02	
2	0	1	32.50	-28.48	1	1	7	25.48	-27.18	8	1	8	3.83	3.65	2	20	5.81	4.55	11	7	4	18.48	-16.87	
2	0	2	71.65	68.42	1	1	8	6.14	-6.08	9	-1	9	9.12	-10.96	2	21	11.69	-8.32	11	7	5	11.87	-11.33	
2	0	3	48.28	48.13	1	1	9	18.85	17.67	9	-1	8	8.43	-9.03	2	22	20.37	-28.41	11	7	6	11.57	10.42	
2	0	4	6.22	-3.41	1	1	10	12.22	13.68	9	-1	7	12.76	13.00	2	23	8.26	-7.79	11	7	7	9.27	7.73	
2	0	5	7.84	-4.87	1	1	11	4.40	-4.51	9	-1	6	4.36	23.14	2	24	9.46	7.80	11	7	8	7.80	-10.85	
2	0	6	17.55	-20.83	2	1	-9	4.97	5.53	9	-1	5	4.30	3.78	2	25	11.33	12.44	11	7	9	5.26	4.47	
2	0	7	5.48	5.46	2	1	-8	20.54	-17.92	9	-1	4	20.52	-17.51	2	26	10.84	10.80	11	7	10	9.20	8.75	
4	0	-10	3.44	-4.88	2	1	-5	24.90	-22.30	9	-1	3	19.68	-18.69	4	2	-10	4.74	-5.37	11	7	-11	3.81	5.88
4	0	-9	16.70	-18.54	2	1	-4	30.22	-28.96	9	-1	2	15.88	12.48	4	3	-11	4.74	-5.79	11	7	-10	4.47	4.25
4	0	-8	21.80	22.29	2	1	-3	25.70	23.26	9	-1	1	4.22	3.28	4	4	-12	5.85	-2.74	11	7	-9	7.64	-6.73
4	0	-7	7.10	7.14	2	1	-2	44.65	48.29	9	-1	0	11.42	-7.66	4	5	-13	5.58	-1.24	11	7	-8	2.94	4.25
4	0	-6	34.52	-34.30	2	1	-1	31.46	28.54	9	-1	-1	23.05	-24.58	4	6	-14	15.50	-15.23	11	7	-7	14.28	14.28
4	0	-5	30.46	30.00	2	1	0	36.46	-63.44	9	-1	0	16.90	17.50	4	7	-15	30.83	-31.10	11	7	-6	15.20	-15.20
4	0	-4	39.19	40.75	2	1	1	14.47	-9.97	9	-1	1	4.36	4.36	4	8	-16	19.07	-18.10	11	7	-5	10.30	-9.04
4	0	-3	28.83	28.40	2	1	2	21.48	21.93	9	-1	2	16.93	17.78	4	9	-17	18.77	-13.26	11	7	-4	5.76	5.29
4	0	-2	37.28	-12.74	2	1	3	22.40	-19.92	9	-1	3	8.44	-13.42	4	10	-18	8.44	-13.42	11	7	-3	6.80	6.80
4	0	-1	26.25	24.93	2	1	4	4.43	5.32	10	-1	8	3.71	3.74	4	11	-19	32.24	32.34	11	7	-2	9.05	-10.35
4	0	0	57.21	-65.22	2	1	5	29.63	-25.52	10	-1	6	5.88	-5.21	4	12	30.13	28.83	11	7	-1	11.55	-11.62	
4	0	1	17.96	-19.08	2	1	6	4.35	4.72	10	-1	5	5.01	4.44	4	13	4.42	2.14	11	7	0	10.17	10.54	
4	0	2	6.25	7.07	2	1	7	12.01	-9.73	10	-1	4	4.38	2.95	4	14	7.38	7.38	11	7	1	5.48	-4.63	
4	0	3	18.78	20.41	2	1	8	10.48	-9.04	10	-1	3	4.15	-0.70	4	15	6.23	-25.81	11	7	2	6.42	-5.74	
4	0	4	12.02	10.48	2	1	9	6.34	-4.22	10	-1	2	2.68	0.80	4	16	-15	12.41	12.41	11	7	3	4.69	-6.71
4	0	5	29.52	-29.99	2	1	10	7.10	-8.56	10	-1	1	8.04	-6.87	4	17	21.72	18.54	11	7	4	4.92	-2.73	
4	0	6	4.75	-4.75	2	1	11	12.21	-11.31	10	-1	0	4.34	3.94	4	18	-14	13.91	13.91	11	7	5	8.01	4.72
4	0	7	33.49	35.35	3	1	-9	8.26	-7.73	10	-1	1	13.73	13.92	5	2	-18	8.88	-8.18	11	7	6	4.10	4.72
4	0	8	14.08	14.94	3	1	-8	5.16	2.37	10	-1	2	8.38	9.34	5	3	-19	6.58	-9.34	11	7	7	3.37	-4.03
4	0	9	7.49	-4.16	3	1	-7	22.65	21.34	10	-1	3	15.18	-16.72	5	4	-20	13.01	-13.01	11	7	8	4.47	4.73
4	0	10	13.44	-11.49	3	1	-6	11.13	10.10	10	-1	4	4.58	-4.02	5	5	-21	5.81	-5.81	11	7	9	6.43	-6.71
6	0	-9	10.52	12.10	3	1	-5	30.47	-29.87	10	-1	5	4.54	3.16	5	6	-22	11.68	-11.68	11	7	10	45.48	-42.30
6	0	-8	5.82	6.13	3	1	-4	35.81	-32.99	11	-1	8	6.06	6.97	5	7	-23	5.58	-11.73	11	7	11	2.94	2.73
6	0	-7	10.29	-9.16	3	1	-3	26.32	27.00	11	-1	7	10.56	9.85	5	8	-24	19.00	-19.00	11	7	12	4.75	13.87
6	0	-6	11.20	11.20	3	1	-2	19.48	-19.54	11	-1	6	10.86	-10.54	5	9	-25	8.06	-8.06	11	7	13	6.48	-11.64
6	0	-5	17.87	14.43	3	1	-1	51.07	54.37	11	-1	5	18.51	22.43	5	10	-26	36.80	-36.80	11	7	14	10.37	-7.33
6	0	-4	49.61	47.08	3	1	0	70.06	-84.02	11	-1	4	16.17	16.18	5	11	-27	19.96	-17.10	11	7	15	8.15	6.34
6	0	-3	18.70	17.64	3	1	1	50.18	-57.44	11	-1	3	13.10	-10.92	5	12	-28	33.82	-33.35	11	7	16	4.57	3.09
6	0	-2	44.20	-48.28	3	1	2	30.52	-30.32	11	-1	2	10.44	-10.44	5	13	-29	20.49	-20.49	11	7	17	8.77	8.77
6	0	-1	34.13	-37.73	3	1	3	29.14	30.08	11	-1	1	2.42	14.30	5	14	-30	9.71	6.88	11	7	18	4.38	-3.46
6	0	0	30.17	29.38	3	1	4	19.34	16.04	11	-1	0	10.84	-7.78	5	15	-31	6.76	4.20	11	7	19	5.21	-1.61
6	0	1	28.46	25.64	3	1	5	24.49	-23.70	11	-1	1	11.84	-11.84	5	16	-32	8.06	-7.16	11	7	20	7.81	-6.22
6	0	2	16.72	-17.82	3	1	6	25.33	-26.40	11	-1	2	3.98	7.21	5	17	-33	4.46	-5.48	11	7	21	9.22	-6.23
6	0	3	40.47	-40.54	3	1	7	9.78	-8.62	11	-1	3	6.43	4.74	6	2	-34	9.27	12.74	11	7	22	7.32	6.24
6	0	4	27.15	-27.04	3	1	8																	

Table 3 (cont.)

4 3 7	5.29	-2.27	0 4 4	14.65	-13.40	9 4 -8	3.93	8.75	6 5 -6	16.41	18.54	6 4 -3	5.16	5.30
4 3 8	12.90	-12.44	0 4 5	18.77	18.26	9 4 -7	7.28	8.42	6 5 -5	20.87	20.10	6 4 -2	8.82	9.41
4 3 9	11.80	-14.98	0 4 6	18.33	17.02	9 4 -6	8.28	-8.83	6 5 -4	12.07	-11.55	6 4 -1	7.17	-8.22
5 3 -10	9.58	-4.72	1 4 0	9.71	-4.41	9 4 -5	9.71	-23.82	6 5 -3	23.82	-22.31	6 4 0	4.45	-3.04
5 3 -9	4.84	6.24	1 4 -8	11.95	9.84	9 4 -4	-14.00	15.65	6 5 -2	9.58	-6.88	6 4 1	6.44	5.85
5 3 -8	7.37	6.96	1 4 -7	27.18	26.33	9 4 -3	6.26	4.66	6 5 -1	14.37	14.15	6 4 2	10.47	8.34
5 3 -7	7.08	-9.20	1 4 -6	36.13	-38.26	9 4 -2	10.03	-8.99	6 5 0	14.33	14.09	6 4 3	4.85	3.73
5 3 -6	10.66	-7.92	1 4 -5	26.27	-33.82	9 4 -1	10.80	-11.34	6 5 1	6.76	-6.76	6 4 4	5.78	9.33
5 3 -5	14.80	14.94	1 4 -4	-18.19	16.74	9 4 0	3.93	-3.76	6 5 2	8.89	-9.52	7 4 -4	7.47	-6.40
5 3 -4	23.76	20.00	1 4 -3	33.96	32.12	9 4 1	7.60	7.08	6 5 3	4.29	-4.10	7 4 -3	7.25	-7.04
5 3 -3	8.90	-5.00	1 4 -2	10.66	-9.36	9 4 2	13.02	13.03	6 5 4	9.56	12.88	7 4 -2	6.66	6.41
5 3 -2	22.98	-18.28	1 4 -1	34.28	-35.46	9 4 3	5.22	5.24	6 5 5	6.53	-6.53	7 4 -1	12.58	11.84
5 3 -1	22.68	-19.74	1 4 0	9.61	-5.97	9 4 4	8.67	-10.05	7 5 -6	2.06	2.52	7 4 0	16.88	-16.17
5 3 0	4.08	3.75	1 4 1	12.41	9.21	10 4 -7	4.16	5.90	7 5 -5	6.12	5.80	7 4 1	16.50	-16.10
5 3 1	18.43	14.48	1 4 2	16.71	14.69	10 4 -6	5.91	-4.12	7 5 -4	11.32	10.35	7 4 2	14.01	15.25
5 3 2	7.54	-6.64	1 4 3	11.07	9.25	10 4 -5	4.44	-8.90	7 5 -3	10.15	-9.58	7 4 3	7.32	8.33
5 3 3	4.54	4.11	1 4 4	5.10	-0.93	10 4 -4	-2.76	12.52	7 5 0	10.71	8.75	8 4 -5	6.08	-6.71
5 3 4	5.61	7.18	1 4 5	9.66	-8.60	10 4 -3	-1.96	8.00	7 5 1	4.92	2.66	8 4 -4	4.25	-3.22
5 3 5	4.97	-4.87	1 4 6	6.90	-8.12	10 4 -2	18.25	-13.34	7 5 2	6.58	-3.81	8 4 -3	7.26	7.00
6 3 -8	7.23	-8.16	1 4 7	8.57	13.55	10 4 -1	15.07	-10.98	7 5 3	8.28	-5.23	8 4 -2	4.40	-1.43
6 3 -7	8.31	8.38	1 4 8	6.64	8.30	10 4 2	12.24	-10.36	8 5 -7	6.21	9.17	8 4 -1	14.37	-13.24
6 3 -6	18.98	16.03	2 4 0	9.78	-10.40	10 4 3	4.20	1.96	8 5 -6	1.87	4.02	8 4 0	12.04	-7.70
6 3 -5	17.03	-13.54	2 4 1	4.15	-4.00	10 4 4	1.82	-4.57	8 5 -5	8.03	-8.20	8 4 1	5.51	7.14
6 3 -4	43.03	-42.64	2 4 2	6.20	5.11	11 4 -6	11.50	-13.87	8 5 -4	16.12	-16.46	8 4 2	9.48	8.84
6 3 -3	20.62	17.58	2 4 3	19.10	18.07	11 4 -5	15.20	-15.29	8 5 -3	10.00	18.74	8 4 3	6.09	-7.08
6 3 -2	10.88	6.23	2 4 4	5.38	5.00	11 4 -4	-8.75	9.10	8 5 -2	14.89	16.52	9 4 -4	4.84	-5.81
6 3 -1	4.51	-2.14	2 4 5	11.07	9.25	11 4 -3	1.82	-4.52	8 5 -1	7.64	-7.64	9 4 -3	7.93	14.65
6 3 0	25.38	-22.86	2 4 6	7.22	7.26	11 4 -2	16.90	-15.14	8 5 0	17.49	-19.58	9 4 -2	14.50	16.30
6 3 1	9.78	-7.77	2 4 7	13.61	-11.83	11 4 0	2.18	2.61	8 5 1	8.68	-8.96	9 4 -1	4.57	4.25
6 3 2	10.86	9.49	2 4 8	2.80	-5.73	11 4 1	11.57	10.41	8 5 2	9.54	9.94	9 4 0	11.07	-13.07
6 3 3	24.93	25.99	2 4 9	3.65	4.81	11 4 2	3.65	4.81	8 5 3	10.49	10.02	9 4 1	9.40	10.02
6 3 4	8.23	-8.84	2 4 0	9.00	-8.21	11 4 3	4.75	-8.33	8 5 4	6.00	-6.50	9 4 2	5.18	6.25
7 3 -9	4.76	-5.84	2 4 1	7.63	-7.62	12 4 -3	5.70	4.63	9 5 0	5.01	-5.07	9 4 3	5.72	8.17
7 3 -8	5.75	-5.95	2 4 2	2.75	3.63	12 4 -2	5.72	5.32	9 5 1	6.90	5.26	10 4 -4	3.54	5.64
7 3 -7	10.25	8.18	2 4 3	8.29	8.21	12 4 -1	-3.00	9.10	9 5 2	-1.06	3.35	10 4 -3	4.80	4.80
7 3 -6	9.41	8.28	3 4 -8	12.73	11.30	12 4 0	4.32	-1.30	9 5 3	11.67	10.34	10 4 0	4.40	-3.41
7 3 -5	8.80	-6.24	3 4 -7	12.49	-9.10	13 4 -4	5.11	4.83	9 5 4	7.95	5.04	10 4 1	2.65	-3.80
7 3 -4	6.93	-3.45	3 4 -6	12.35	-12.60	13 4 -3	7.44	-4.73	9 5 5	7.01	-4.73	10 4 2	10.17	-11.29
7 3 -3	8.87	7.54	3 4 -5	23.10	22.12	13 4 -2	14.11	-13.44	9 5 6	4.55	-3.20	10 4 3	6.14	14.94
7 3 -2	20.39	18.70	3 4 -4	36.19	32.74	13 4 0	12.61	13.35	9 5 7	4.16	6.30	10 4 4	13.81	14.90
7 3 -1	4.20	1.90	3 4 -3	19.00	-16.58	13 4 1	8.26	7.71	10 5 -6	5.20	-6.60	10 4 5	9.16	10.41
7 3 0	13.98	-18.60	3 4 -2	41.76	-41.84	13 4 2	6.92	-1.78	10 5 -5	7.83	-5.78	1 7 -4	11.31	-15.69
7 3 1	17.38	-15.29	3 4 0	23.51	-22.08	13 4 3	7.74	-10.13	10 5 -4	14.26	12.85	1 7 -5	8.03	-8.55
7 3 2	11.50	-9.07	3 4 1	14.55	14.65	14 4 0	3.46	2.31	10 5 -3	2.77	3.10	1 7 -6	12.08	12.85
7 3 3	8.82	7.70	3 4 2	25.86	24.91	14 4 1	3.40	4.38	10 5 -2	15.65	-15.55	1 7 -7	14.96	13.82
7 3 4	11.50	8.50	3 4 3	14.50	-12.55	14 4 2	14.50	-12.55	10 5 -1	15.65	-15.55	1 7 -8	14.96	13.82
8 3 -9	3.56	-4.36	3 4 4	5.14	-14.44	0 5 2	16.91	-13.58	10 5 0	7.15	6.76	1 7 0	5.50	-4.78
8 3 -8	3.66	2.11	3 4 5	6.64	-5.58	0 5 3	17.81	17.32	10 5 1	2.07	19.91	1 7 1	4.33	3.80
8 3 -7	15.74	15.91	3 4 6	13.61	14.14	0 5 4	17.30	15.87	10 5 2	7.94	6.89	1 7 2	7.30	-9.09
8 3 -6	14.22	18.59	3 4 7	14.22	11.28	0 5 5	12.20	11.28	10 5 3	12.91	11.89	1 7 3	4.11	-7.46
8 3 -5	5.82	-5.44	3 4 8	2.93	-4.08	0 5 6	8.78	-8.17	10 5 4	5.38	-4.89	1 7 4	8.36	9.92
8 3 -4	9.81	-5.68	4 4 -9	4.30	4.90	0 5 7	16.44	-17.02	11 5 -5	3.61	-8.81	2 7 -2	7.42	7.05
8 3 -3	7.30	-3.86	4 4 -8	5.14	-5.16	1 5 -8	6.39	5.39	11 5 -4	7.47	-5.56	2 7 -1	8.77	9.92
8 3 -2	15.73	12.44	4 4 -7	2.42	4.44	1 5 -7	3.42	4.26	11 5 -3	5.45	-3.72	2 7 0	4.87	-14.01
8 3 -1	13.39	12.17	4 4 -6	14.94	-14.11	1 5 -6	12.51	-11.49	11 5 2	6.12	3.31	2 7 1	8.46	-7.16
8 3 0	26.79	-27.76	4 4 -5	7.57	-9.43	1 5 -5	4.89	-5.80	11 5 1	7.28	4.45	2 7 2	10.07	10.28
8 3 1	20.80	-17.48	4 4 -4	11.26	8.28	1 5 -4	2.56	0.87	12 5 -2	8.45	-9.74	2 7 3	9.73	11.33
8 3 2	5.40	8.80	4 4 -3	11.26	9.21	1 5 -3	4.42	-2.72	12 5 -1	8.45	-9.74	2 7 4	8.52	9.77
8 3 3	9.51	8.99	4 4 -2	23.02	22.17	1 5 -2	1.41	3.43	12 5 0	1.65	3.78	2 7 5	3.46	-5.93
8 3 4	7.35	6.09	4 4 -1	3.09	2.98	1 5 0	6.85	4.98	12 5 1	9.56	11.93	3 7 -6	4.70	-6.17
8 3 5	12.90	-12.44	4 4 0	12.49	-10.67	1 5 1	4.91	3.61	12 5 2	3.40	3.79	3 7 -5	6.87	5.78
8 3 6	11.09	-9.65	4 4 1	14.70	13.39	1 5 2	11.90	15.51	12 5 3	-1.40	-3.73	3 7 -4	4.92	-3.73
9 3 -8	7.96	-8.61	4 4 2	7.74	6.41	1 5 3	19.15	17.77	0 6 0	24.04	-22.18	3 7 -3	15.36	-14.60
9 3 -7	7.89	8.47	4 4 3	10.02	-8.27	1 5 4	6.95	7.67	0 6 1	4.77	-4.45	3 7 -2	14.25	15.00
9 3 -6	14.50	13.31	4 4 4	12.56	-11.50	1 5 5	12.56	-11.50	0 6 2	11.50	6.92	3 7 -1	11.58	5.95
9 3 -5	7.81	8.53	4 4 5	4.00	3.91	1 5 6	11.92	-9.98	0 6 3	7.07	7.12	3 7 0	10.15	-10.07
9 3 -4	16.84	-14.80	4 4 6	2.73	-4.62	2 5 -8	5.97	7.01	0 6 4	7.08	5.20	3 7 1	5.45	5.41
9 3 -3	21.82	-21.84	4 4 7	8.33	-8.83	2 5 -7	7.10	6.04	0 6 5	9.54	10.43	4 7 -5	8.55	8.85
9 3 -2	18.90	-19.35	4 4 8	8.90	-9.35	2 5 -6	11.92	11.92	0 6 6	10.50	10.50	4 7 -4	12.95	16.14
9 3 -1	18.08	17.64	4 4 9	15.21	15.38	2 5 -5	18.72	-18.46	1 6 -4	4.67	-10.63	4 7 -3	5.73	8.32
9 3 0	25.93	-15.21	4 4 0	10.59	9.84	2 5 -4	6.96	5.66	1 6 -3	10.13	9.86	4 7 -2	16.47	-8.75
9 3 1	11.18	8.97	4 4 -3	9.36	6.85	2 5 -3	17.11	17.74	1 6 -2	23.50	24.53	4 7 -1	14.47	-14.54
9 3 2	14.50	-15.17	4 4 -2	15.41	-11.06	2 5 -2	27.90	-22.55	1 6 -1	27.90	-22.55	4 7 0	7.32	10.60
9 3 3	7.10	-8.91	4 4 -1	11.35	-9.78	2 5 -1	2.70	0.50	1 6 0	19.19	-18.24	4 7 1	8.28	-9.43
10 3 -8	3.73	4.65	4 4 0	24.49	24.39	2 5 0	37.07	-40.28	1 6 1	18.26	-20.70	4 7 2	10.60	-12.34
10 3 -7	3.51	-6.35	4 4 1	15.09	14.39	2 5 1	17.57	-17.53	1 6 2	19.21	21.00	4 7 3	5.38	-6.70
10 3 -6	18.82	-16.72	4 4 2	18.82	-16.72	2 5 2	19.85	-17.95	1 6 3	16.85	13.95	4 7 4	6.33	7.94
10 3 -5	10.83	-8.98	4 4 3	23.05	-23.81	2 5 3	23.86	25.31	1 6 4	5.10	-10.44	4 7 5	11.82	10.31
10 3 -4	4.25	3.90	4 4 4	8.41	-7.42	2 5 4	4.42	2.72	1 6 5	15.23	-19.03	4 7 6	2.21	-2.07
10 3 -3	24.35	13.96	4 4 5	13.96	24.35	2 5 5	23.89	-22.48	1 6 6	13.96	24.35	4 7 7	13.94	-18.24
10 3 -2	19.41	18.27	4 4 6	9.56	8.87	2 5 6	5.29	-3.93	1 6 7	5.80	-6.21	4 7 8	6.25	6.08
10 3 -1	5.63	-3.71	4 4 7	4.82	-3.26	2 5 7								

Table 4. Bond lengths and angles of the proline molecule with their standard deviations

C(1)–O(1)	1.239 ± 0.012 Å	O(1)–C(1)–O(2)	124.5 ± 0.9°
C(1)–O(2)	1.323 ± 0.012	O(1)–C(1)–C(2)	122.7 ± 0.8
C(1)–C(2)	1.519 ± 0.013	O(2)–C(1)–C(2)	112.9 ± 0.8
C(2)–C(3)	1.543 ± 0.013	C(1)–C(2)–N	111.3 ± 0.7
C(3)–C(4)	1.506 ± 0.020	C(1)–C(2)–C(3)	113.0 ± 0.8
C(4)–C(5)	1.504 ± 0.016	N—C(2)–C(3)	106.8 ± 0.7
C(5)–N	1.516 ± 0.014	C(2)–C(3)–C(4)	100.8 ± 0.8
N—C(2)	1.473 ± 0.011	C(3)–C(4)–C(5)	109.2 ± 1.1
		C(4)–C(5)–N	106.0 ± 1.0
		C(5)–N—C(2)	104.6 ± 0.7

standard scale. These intensities were corrected for Lorentz and polarization factors in the usual way, but no absorption correction was applied. The sets of intensities for the two axes were scaled by correlating the common reflexions. A total of 1239 independent observed structure factors, out of the theoretically allowed 1650 reflexions in the limiting sphere for Cu  $K\alpha$  radiation, were obtained, and they were put on an absolute scale by Wilson's (1942) statistical method.

#### Determination and refinement of the structure

A three-dimensional sharpened Patterson synthesis yielded the coordinates of the chlorine atom. A three-dimensional electron density map with signs determined by the chlorine atom showed seven out of the nine atoms in an asymmetric unit. The second electron density map, however, showed all nine atoms and this structure was confirmed by the third Fourier synthesis. The refinement was first carried out by five cycles of the block-matrix least-squares calculations with isotropic temperature factors. The  $R$  value reduced from 0.27 to 0.15. At this stage, five cycles of the full-matrix least-squares refinement with the *ORFLS* program (Busing, Martin & Levy, 1962) was started taking account of anisotropic thermal vibrations for each atom. In this

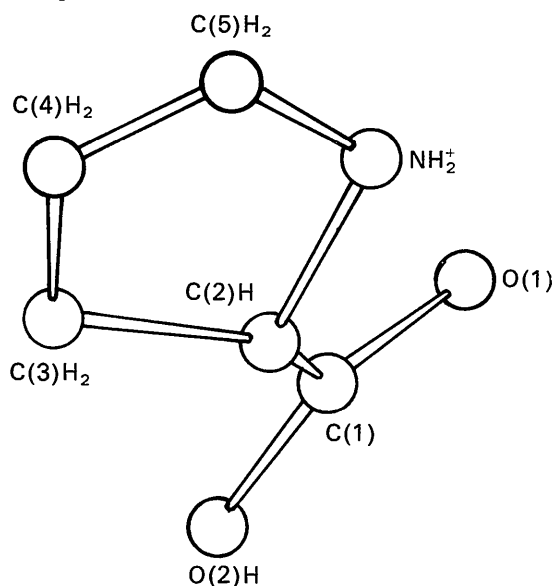


Fig. 1. L-Proline molecule in DL-proline hydrochloride.

calculation, the following weighting scheme was adopted:

$$\begin{aligned} \sqrt{w} &= F_o/20, & \text{when } F_o \leq 20, \\ \sqrt{w} &= 20/F_o, & \text{when } 20 < F_o < 80, \\ \sqrt{w} &= 20 \times 80/F_o^2, & \text{when } 80 \leq F_o. \end{aligned}$$

The  $R$  values in the last three cycles were all around 0.124 excluding hydrogen atom contributions. The positional parameter shifts at the final cycles were less than 0.00024 Å with an average value of 0.00008 Å, except those of the  $x$  and  $y$  coordinates of C(4) and C(1) and of the  $z$  coordinates of C(5) and C(1), for which the maximum shift calculated was 0.00064 Å and the average shift 0.00050 Å.

The atomic scattering factors used were those given by Berghuis, Haanappel, Potters, Loopstra, MacGillavry & Veenendaal (1955) for carbon, oxygen and nitrogen atoms, and that given by Dawson (1960) for the chloride ion.

The final atomic parameters and their standard deviations are listed in Table 1. Table 2 shows the final anisotropic thermal parameters. Observed and calculated structure factors are listed in Table 3.

#### Description of the structure

Fig. 1 shows the structure of the L-proline molecule. Intramolecular bond lengths and angles are listed in Table 4 together with their standard deviations. In Table 5 the internal rotation angles of the L-molecule are shown. This angle for a series of atoms  $A-B-C-D$  is defined as follows: it is the angle formed by the projection of the  $A-B$  bond with that of  $C-D$  when the projection is taken along the  $B-C$  bond; the angle is taken as positive if the rotation to bring the projection of  $A-B$  into that of  $C-D$  would coincide with that of the right handed screw advancing along the  $B-C$  bond.

Table 5. Internal rotation angles of the proline molecule (L-form)

O(1)–C(1)–C(2)–C(3)	131.1°
O(2)–C(1)–C(2)–N	–167.6
O(2)–C(1)–C(2)–C(3)	–47.4
O(1)–C(1)–C(2)–N	11.0
C(1)–C(2)–C(3)–C(4)	–157.7
C(2)–C(3)–C(4)–C(5)	23.0
C(3)–C(4)–C(5)–N	–3.8
C(4)–C(5)–N—C(2)	–18.4
C(5)–N—C(2)–C(1)	157.4
C(3)–C(2)–N—C(5)	33.7
C(4)–C(3)–C(2)–N	–35.0

The root-mean-square displacement of the atoms along the three principal axes of the thermal ellipsoids and the angles between each principal axis and the three cartesian axes are listed in Table 6. The latter

axes are related to the crystallographic axes as follows: axes 1 and 3 are parallel to **a** and **b** respectively, and axis 2 is defined in such a way that it completes a right handed system with the other two axes. To illustrate

Table 6. Root mean square (r.m.s.) components of thermal vibrations along three principal axes and angles between these and the cartesian axes

For definition of the latter axes, see text.

	Principal axis	R.m.s. component	Angles with cartesian axes		
			1	2	3
Cl	1	0.128 Å	61°	90°	29°
	2	0.165	126	42	71
	3	0.203	49	48	111
C(1)	1	0.112	42	117	60
	2	0.167	111	69	31
	3	0.214	55	35	96
C(2)	1	0.135	48	110	131
	2	0.156	138	102	129
	3	0.172	86	24	114
C(3)	1	0.101	87	10	81
	2	0.175	138	93	48
	3	0.307	48	99	43
C(4)	1	0.122	117	49	53
	2	0.193	125	137	67
	3	0.479	47	101	45
C(5)	1	0.125	71	123	141
	2	0.217	148	122	88
	3	0.289	115	50	129
N	1	0.116	42	93	132
	2	0.144	123	129	124
	3	0.176	113	39	120
O(1)	1	0.155	102	52	41
	2	0.208	134	130	70
	3	0.232	46	117	56
O(2)	1	0.138	41	69	124
	2	0.199	89	149	121
	3	0.228	47	111	49

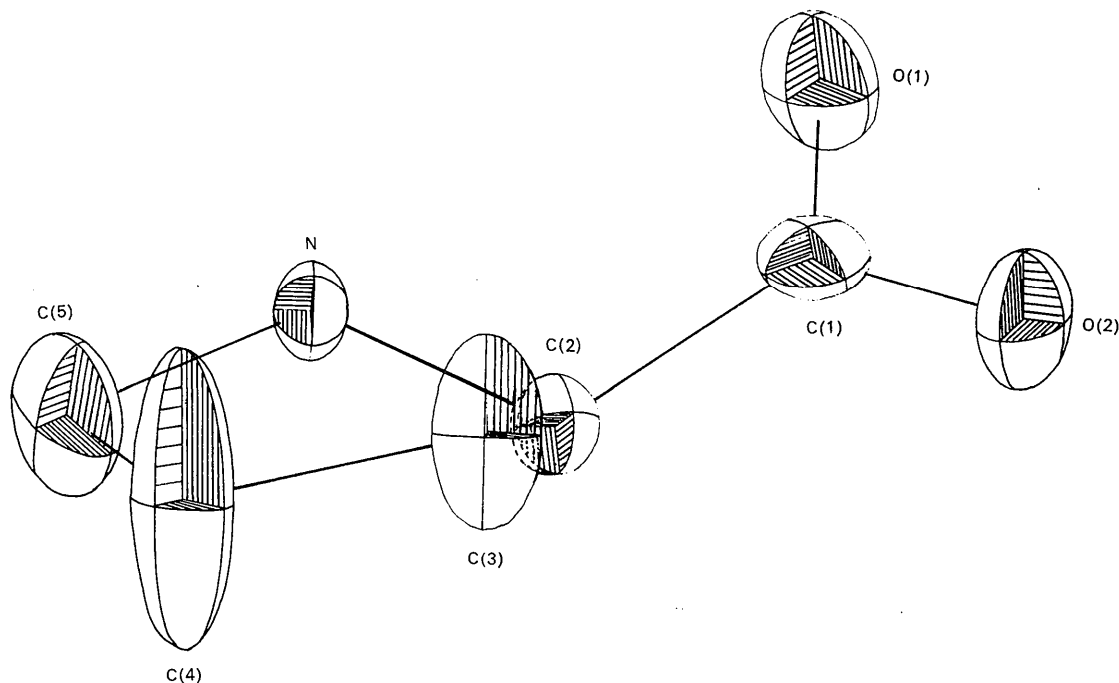


Fig. 2. The vibrational ellipsoids for atoms of the D-proline molecule.

the relation between the directions of the principal axes of thermal vibration and the molecular structure, the vibration ellipsoids are shown in Fig. 2. This Figure was drawn by the plotter program *ORTEP* (Johnson, 1965). The area covered by the ellipsoid is such that the probability of finding the centre of the atom in it is 50%.

The nitrogen atom seems to be in the form of  $>NH_2^+$ , as indicated by the hydrogen bond scheme described later. The carboxyl group atoms C(2), C(1), O(1) and O(2) are coplanar, with a maximum deviation of 0.008 Å. The bond lengths of C(1)–O(1) and C(1)–O(2) indicate that the hydrogen atom is attached to O(2).

The conformation of the pyrrolidine ring is unusual: four atoms [C(3), C(4), C(5) and N] are coplanar within 0.02 Å, while the fifth atom, C(2), is 0.5 Å away from this plane. In all six cases of related compounds [L-proline (Kayushina & Vainshtein, 1965), hydroxyproline (Zussman, 1951; Donohue & Trueblood, 1952), copper proline (Mathieson & Welsh, 1952) the prolyl and hydroxyprolyl residues in tosyl-prolylhydroxyproline (Fridrichsons & Mathieson, 1962) and in leucylprolylglycine (Leung & Marsh, 1958)] the atom C(4) (of the present numbering system) was found to be about 0.5 Å away from the best plane formed by the remaining four atoms.

#### Discussion of the molecular structure

As shown in Fig. 3(a), the nitrogen atom lies nearly in the plane of the carboxyl group, the dihedral angle between N–C(1)–C(2) and the carboxyl plane being 11.0°. This tendency seems to be a common feature of the proline group. Thus, in L-proline the nitrogen atom is 0.23 Å from the carboxyl plane, whereas in hydroxyproline it is only 0.05 Å. In leucylprolylglycine, though in this case the proline residue forms a peptide bond, the corresponding dihedral angle is about 17°. The only exception is found at the terminal carboxyl group in tosyl-prolylhydroxyproline, where the carboxyl plane makes an angle of about 90° to the plane of the pyrrolidine ring.

The puckering of the pyrrolidine ring is the subject to be considered next. From the study on cyclopentane (Pitzer, 1945; Kilpatrick, Pitzer & Spitzer, 1947), the puckering of the pyrrolidine ring seems to be a result

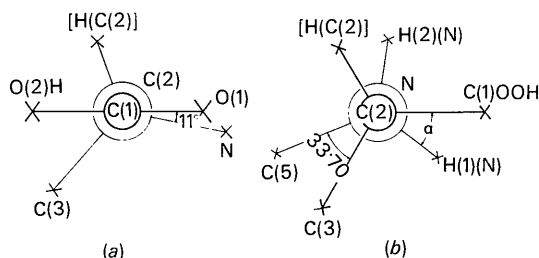


Fig. 3. Internal rotation relationship around the axes (a) C(1)–C(2) and (b) C(2)–N.

of the repulsion of non-ring atoms bonded to adjoining ring-atoms, which would result from the planar structure, since in the planar structure these non-ring atoms are forced to occupy an eclipsed position. In the present case, the puckering of the atom C(2), which has a bulky carboxyl group, would provide more relief than that of other ring atoms having only hydrogen atoms. To make this point clearer, the conformation around the C(2)–N bond is shown in Fig. 3(b). (Note that the carboxyl group lies nearly perpendicular to the surface of the paper.) If it is first assumed that the conformation around C(1)–C(2) is fixed as Fig. 3(a), the following explanation of the origin of the puckering is possible. First, we consider the usual case in which C(4) is puckered. Since C(3)–C(2)–N–C(5) are coplanar in this case, the carboxyl group and H(1) attached to N would be eclipsed. This would make H(1) approach O(1) at a distance as short as 2.2 Å (about 0.4 Å shorter than the sum of normal van der Waals radii). To avoid this short contact of H(1) and O(1), the group H(1), H(2) and C(5) should be rotated around the C(2)–N axis to increase the angle  $\alpha$ , and this inevitably leads to the puckering of C(2) as is observed in the present structure. In the case of hydroxyproline (Donohue & Trueblood, 1952) and L-proline (Kayushina & Vainshtein, 1965), on the other hand, the apparently unfavourable eclipsed conformation described above does occur and this results in the puckering of C(4) instead of C(2), where the internal rotation angles of C(3)–C(2)–N–C(5) are  $-5.2^\circ$  and  $-13.7^\circ$  respectively, instead of  $33.7^\circ$  as in the present structure. In these two cases, one of the ionized carboxyl oxygen atoms is situated at a distance as short as about 2.1 Å from the positively charged imino hydrogen atom and it seems that the electrostatic attraction between these atoms stabilizes the eclipsed conformation. Fig. 4(a), (b) and (c) shows the stereographic projections indicating the relationship between the angles of internal rotation around the C(2)–N bonds and the directions of the intramolecular N...O(1) vectors. The O(1)...H distances are also shown in these Figures. The hydrogen atoms in DL-proline hydrochloride are at the assumed positions. In these Figures the directions of the intermolecular N...O or N...Cl vectors are also shown with the hydrogen bond distances. The strong hydrogen bonds would reduce the internal rotation angle, C(3)–C(2)–N–C(5) in hydroxyproline, while in DL-proline hydrochloride they seem to stabilize the present conformation. On the basis of the data available at present, it may be said that pyrrolidine ring is rather flexible and its conformation seems to be affected by various factors including intermolecular interactions.

The principal-axis analysis of the temperature factor ellipsoids revealed that the atoms C(3), C(4), C(5) and especially C(4) have an unusually large vibrational amplitude in the direction roughly perpendicular to the best plane of the pyrrolidine ring (Table 6 and Fig. 2). This may be interpreted by consideration of one of the following alternatives: (1) puckering vibration of C(4)

with two equilibrium positions on both sides of the plane; (2) simple but non-localized vibration such as the oscillatory vibration of the plane C(3)-C(4)-C(5). In an analysis of leucylprolylglycine, Leung & Marsh (1958) assigned the two 'half atoms' to C(4) (their C<sub>6</sub>) of the pyrrolidine ring, and the puckering of the ring seems to occur mainly at C(4) rather than at other posi-

tions. In this case, the difference electron density map based on the model in which a single atom is put at the average position of the two 'half atoms' showed two predominant ( $1.0 \sim 1.25 \text{ e. \AA}^{-3}$ ) peaks on either side of the assumed position of C(4). These peaks remained even after the anisotropic temperature factor was applied to  $F_o$ . In view of these facts, we calculated the dif-

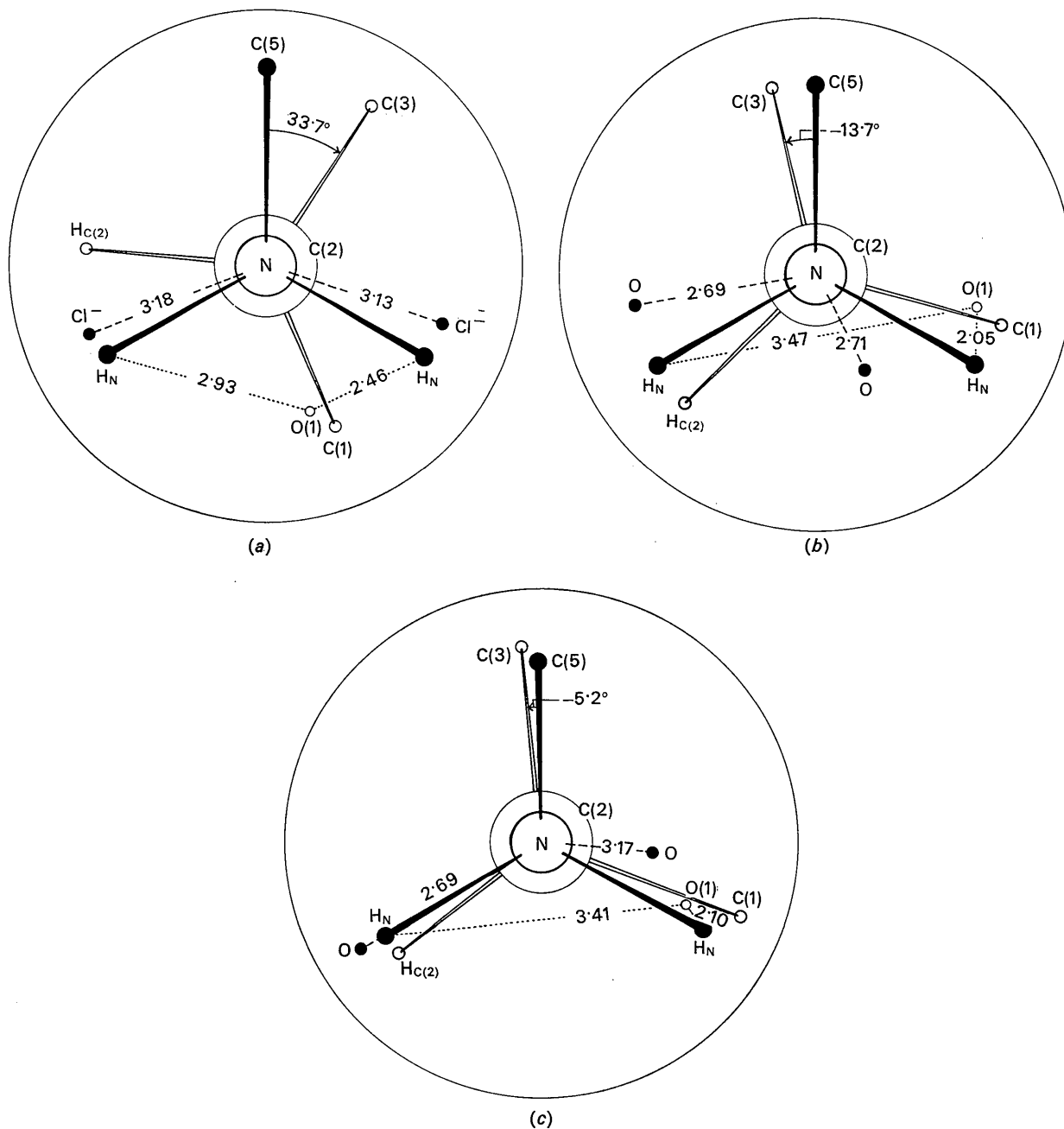


Fig.4. Stereographic projection showing the arrangement of oxygen and chlorine atoms around the nitrogen atom. The direction of the C(2)-N bond is taken as a polar axis. The interatomic distances are also shown. Broken lines indicate hydrogen bonds. (a) DL-Proline hydrochloride (present study). (Note that O(1) is a carbonyl oxygen atom.) (b) L-Proline (Kayushina & Vainshtein, 1965). (c) L-Hydroxyproline (Donohue & Trueblood, 1952).

ference Fourier synthesis with the final atomic coordinates and anisotropic temperature factors. No significant peak (more than  $0.3 \text{ e.}\text{\AA}^{-3}$ ) appeared in the vicinity of C(3), C(4) and C(5). This, however, does not necessarily exclude the possibility of puckering. In the usual least-squares refinement, although a thermal vibration with only one equilibrium position is assumed, a puckering vibration with two equilibrium positions may well be approximated by an apparent vibrational ellipsoid which is unusually elongated in the puckering direction. The following point, however, seems to favour the second alternative. Two atoms adjacent to C(4), namely C(3) and C(5), also have large amplitudes perpendicular to the best plane of the pyrrolidine ring. This may be best explained by assuming out-of-plane vibrations in which the atoms C(3) and C(5) are vibrating either in phase or  $180^\circ$  out of phase with respect to C(4). The latter mode of vibration involves only deformation of the internal rotation angles within the pyrrolidine ring and it seems to be very plausible. However, the former mode consisting of an oscillatory vibration of the C(3)–C(4)–C(5) plane as a rigid body

around the axis lying parallel and near to the C(2)–N bond would also be possible, probably as part of the librating motion of the molecule and this accounts well for the magnitudes of the root mean square displacements of the largest vibrations of the three atoms. There is always another possibility, that the statistical distribution of various distorted molecules over the crystal also results in the apparently large 'temperature factor'. Distinguishing between this and the real vibration is beyond the power of the present method.

Finally, it must be mentioned that such a large movement of atoms tends to give a bond length shorter than the true value (Busing & Levy, 1964). The slightly short C–C distances involving C(4), namely  $1.506 \text{ \AA}$  of C(3)–C(4) and  $1.504 \text{ \AA}$  of C(4)–C(5), might be a result of this effect.

In view of the importance of deducing standard structural parameters for the proline residue, the bond lengths and angles determined in the present study are compared with those of five other analyses in Table 7. For all but copper proline the parameters were determined by three-dimensional analysis; among these,

Table 7. Comparison of bond lengths and angles found in the compounds of proline and hydroxyproline

Compound Method Residue	DL-Pro-	L-Proline	Hydroxy-	L-Leucyl-	Tosyl-L-prolyl-		Copper
	line. HCl 3D* LSQ	3D LSQ	L-proline 3D LSQ	L-prolyl- glycine 3D LSQ	L-hydroxyproline. H <sub>2</sub> O 3D F	hydroxyprolyl	pro- line. 2H <sub>2</sub> O 2D
Bond lengths (Å)							
C(1)–O(1)	1.239	1.28	1.254	1.236	1.24	1.19	1.24
C(1)–O(2)	1.323	1.26	1.269	—	—	1.41	1.24
C(1)–C(2)	1.519	1.52	1.516	1.519	1.51	1.52	1.50
C(2)–C(3)	1.543	1.52	1.532	1.497	1.56	1.53	1.52
C(3)–C(4)	1.506	1.54	1.503	1.512	1.56	1.54	1.50
C(4)–C(5)	1.504	1.53	1.524	1.504	1.56	1.54	1.52
C(5)–N	1.516	1.48	1.482	1.458	1.50	1.43	1.53
C(2)–N	1.473	1.53	1.503	1.452	1.49	1.45	1.52
Estimated error STD	0.014	0.012	0.009	0.015			—
LE			0.03	0.03		0.06	
Bond angles (°)							
O(1)–C(1)–O(2)	124.5	120	126.1	—	—	124.7	122
O(1)–C(1)–C(2)	122.7	119	118.5	121.4	—	123.4	120
O(2)–C(1)–C(2)	112.9	121	115.4	—	—	140.8	118
C(1)–C(2)–N	111.3	107	110.8	111.2	109.3	117.6	108
C(1)–C(2)–C(3)	113.0	112	113.3	113.3	100.7	114.7	112
N—C(2)–C(3)	106.8	106	104.5	103.7	98.8	103.7	108
C(2)–C(3)–C(4)	100.8	101	107.6	106.9	115.2	103.0	97
C(3)–C(4)–C(5)	109.2	102	103.9	105.7	95.1	106.6	109
C(4)–C(5)–N	106.0	107	105.5	103.4	107.8	98.3	96
C(5)–N—C(2)	104.6	107	109.4	113.3	111.6	116.2	108
Estimated error STD	0.8	0.8		1.0		—	—
LE			1.0	2.0			
State of N	NH <sub>2</sub> <sup>+</sup>	NH <sub>2</sub> <sup>+</sup>	NH <sub>2</sub> <sup>+</sup>	NH–CO–	NH–SO <sub>2</sub>	NH–CO–	NH···Cu
State of carboxyl	–COOH	–COO <sup>–</sup>	–COO <sup>–</sup>	–CO–NH–	–CO–NH–	–COOH	–COO···Cu
Reference	(1)	(2)	(3)	(4)		(5)	(6)
	(1) Present study			(2) Kayushina & Vainshtein (1965)			
	(3) Donohue & Trueblood (1952)			(4) Leung & Marsh (1958)			
	(5) Fridrichsons & Mathieson (1962)			(6) Mathieson & Welsh (1952)			

\* Abbreviations: 2D, two-dimensional analysis; 3D, three-dimensional analysis; LSQ, least-squares method; F, Fourier method; STD, standard deviation; LE, limit of error.



parameters of hydroxyproline and leucylprolylglycine were refined by least-squares methods, while those of tosyl-prolylhydroxyproline were obtained by Fourier methods.

### Crystal structure

The  $b$  and  $c$  axis projections are shown in Figs. 5 and 6, respectively. The positions of the molecules are as follows: I( $x, y-1, z$ ); II( $\bar{x}+\frac{1}{2}, y-\frac{1}{2}, \bar{z}+1$ ), III( $\bar{x}+1, \bar{y}+2, \bar{z}+1$ ); IV( $x+\frac{1}{2}, \bar{y}+\frac{3}{2}, z$ ), with the  $x, y$  and  $z$  coordinates as given in Table 1. The subscript attached to the molecular number indicates translations along the three edges of the unit cell. Molecule I is drawn as the D-molecule. The packing scheme may be described as follows: a series of molecules, ...I<sub>001</sub>, Cl, IV<sub>001</sub>, Cl, I<sub>101</sub>..., form a zigzag chain extending along the  $a$  axis, connected by hydrogen bonds No. 1 and No. 3 (Table 9); another series of molecules, ...II, Cl, III, Cl, II<sub>100</sub>..., form the same type of chain. The two chains are connected by hydrogen bonds of type No. 2 to form a sheet extending parallel to (001). In addition to these hydrogen bonds there are many van der Waals contacts between the molecules in the sheet. All these contacts less than 4 Å are listed in Table 8. In contrast to the close contacts within the sheet, the packing of the sheets is surprisingly loose. There is no hydrogen bond and only one kind of contact less than 4 Å is found between Cl(I) and C(4)(II). This fact explains the (001)

cleavage mentioned earlier and to some extent the unusually large temperature factors observed for atoms C(3), C(4) and C(5).

Table 8. Packing distances less than 4 Å

From molecule I	To atom	Of molecule	Translation	Distance	
N	C(5)	II	00 $\bar{1}$	3.86 Å	
O(1)	Cl	I	}	000	3.44
	Cl	IV		3.94	
	Cl	II		00 $\bar{1}$	3.77
	C(5)	I		010	3.62
	C(5)	II		00 $\bar{1}$	3.19
O(2)	N	II	}	00 $\bar{1}$	3.08
	C(2)	III		3.62	
	C(1)	III		3.16	
	O(1)	III		0 $\bar{1}\bar{1}$	3.37
	O(2)	III		3.39	
	C(3)	III		0 $\bar{1}0$	3.39
	C(5)	IV		0 $\bar{1}0$	3.36
	C(1)	III		0 $\bar{1}\bar{1}$	3.89
	O(1)	III		0 $\bar{1}\bar{1}$	3.39
	C(1)	Cl		I	}
Cl		IV	3.84		
C(1)		III	3.43		
O(1)		III	0 $\bar{1}\bar{1}$	3.16	
O(2)		III	3.89		
C(2)	Cl	I	}	000	3.91
	Cl	IV		0 $\bar{1}0$	3.58
	Cl	II		3.84	
	O(1)	III		0 $\bar{1}\bar{1}$	3.62
	O(2)	III		0 $\bar{1}0$	3.39
C(3)	O(2)	III	}	3.96	
	Cl	IV		3.96	

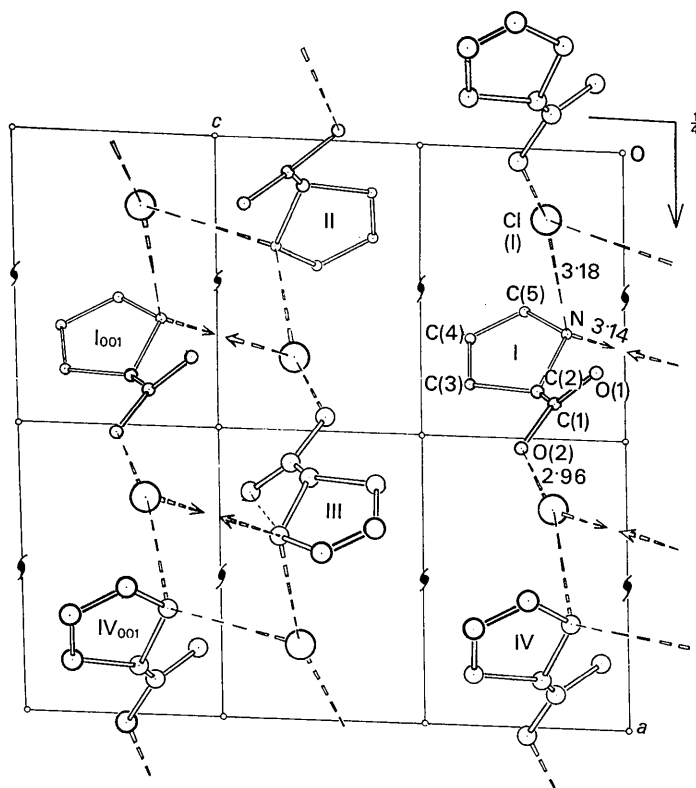


Fig. 5. Projection of the crystal structure along the  $b$  axis. Hydrogen bonds are indicated by single or double broken lines.

Table 8 (cont.)

From molecule I	To atom	Of molecule	Translation	Distance
C(4)	Cl	II	000	3.76
	Cl	II	0 $\bar{1}$ 0	3.76
C(5)	Cl	I	000	3.79
	Cl	I	} 0 $\bar{1}$ 0	3.80
	O(1)	I		3.62
	O(2)	IV	$\bar{1}\bar{1}$ 0	3.36
	Cl	II	} 0 $\bar{1}\bar{1}$	3.84
	N	II		3.86
	O(1)	II		3.19

## Operations

I	$x,$	$y-1,$	$z$
II	$\bar{x}+\frac{1}{2},$	$y-\frac{1}{2},$	$\bar{z}+1$
III	$\bar{x}+1,$	$\bar{y}+2,$	$\bar{z}+1$
IV	$x+\frac{1}{2},$	$\bar{y}+\frac{3}{2},$	$z$

Table 9. *Hydrogen bond distances and angles*

Atoms belong to molecule I unless otherwise specified.

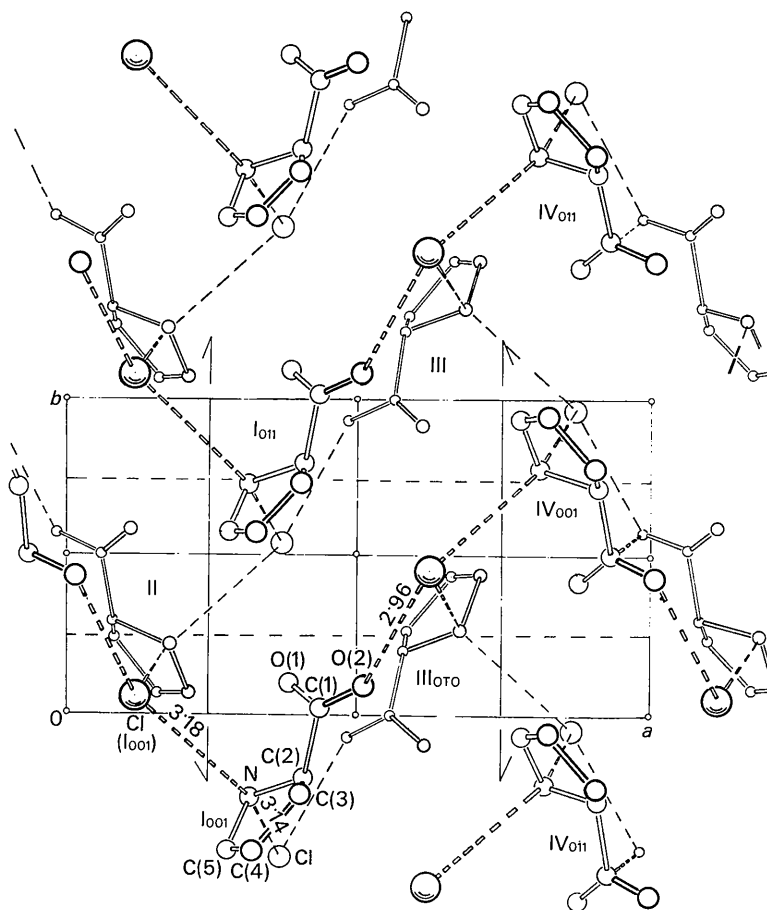
No.		Distance
1	O(2)-H...Cl (IV)	2.958 $\pm$ 0.007 Å
2	N-H...Cl (II $_{0\bar{1}\bar{1}}$ )	3.135 $\pm$ 0.008
3	N-H...Cl	3.180 $\pm$ 0.007

Table 9 (cont.)

	Angle
C(1)-O(2)...Cl	122.4 $\pm$ 1.9°
C(2)-N...Cl	108.7 $\pm$ 0.5
C(5)-N...Cl	101.8 $\pm$ 0.5
Cl (II $_{0\bar{1}\bar{1}}$ )...N...Cl	126.7 $\pm$ 0.2
C(2)-N...Cl (II $_{0\bar{1}\bar{1}}$ )	107.2 $\pm$ 0.5
C(5)-N...Cl (II $_{0\bar{1}\bar{1}}$ )	105.8 $\pm$ 0.6

## Hydrogen bonds

Hydrogen bond distances and angles are listed in Table 9. All three hydrogen bond distances are close to the lower limit of their usual range. This is quite natural considering that they involve the ionized group  $>NH_2^+$  and the ionized atom  $Cl^-$ . The hydrogen bond O(2)-H...Cl(IV) seems to be, within experimental error, completely linear as suggested by the angle C(1)-O(2)...Cl listed in Table 9. The hydrogen bond scheme around the nitrogen atom is shown in Fig. 4 by use of the stereographic projection. Both acceptor Cl atoms lie nearly in tetrahedral directions and we can conclude that the nitrogen atom is in the form  $>NH_2^+$ . In addition,

Fig. 6. Projection of the crystal structure along the  $c$  axis.

tion, there are two other short contacts, namely N...O(1) ( $\Pi_{0II}$ ), 3.08<sub>4</sub> Å and N...O(1), 2.75<sub>2</sub> Å. The atom O(1) belongs to the same molecule while the atom O(1) ( $\Pi_{0II}$ ) lies approximately on the line C(2)–N. The arrangement of this latter type is very similar to that of carbonyl oxygen [O(1'')] around the –NH<sub>3</sub><sup>+</sup> group in valine hydrochloride (Parthasarathy, 1966) or in  $\gamma$ -glycine (Iitaka, 1961).

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## Least-Squares Refinement of the Crystal and Molecular Structures of DNA and RNA from X-ray Data and Standard Bond Lengths and Angles

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A method is described for producing molecular models (with standard bond lengths and angles) that optimally fit the low resolution X-ray diffraction data characteristically obtained from crystalline fibres. The application of this linked-atom, least-squares method in the production of refined structures for DNA and RNA complementary helices is discussed. Atomic coordinates and molecular parameters are presented for these new standard polynucleotide models that not only have contemporary bonded stereochemistry but fit the X-ray intensities to a substantially better degree. The geometrical properties of these new models are analysed in detail and compared with one another and with the conformations found in monomers. In addition, the results of the refinements allow a decision to be made that the RNA helices are eleven- and not tenfold.

### Introduction

The alkali metal salts of native, double-helical polynucleotides exist in a number of different, fully crystalline forms. In the *A* and *B* forms of deoxyribonucleic acid (DNA) the polyanions have very different conformations (also called *A* and *B*) while in the  $\alpha$  and  $\beta$  forms of ribonucleic acid (RNA) the molecular conformations are probably the same. Diffraction analyses of

all four structures have been published (Fuller, Wilson, Wilkins, Hamilton, & Arnott, 1965; Langridge, Marvin, Seeds, Wilson, Hooper, Wilkins & Hamilton, 1960; Arnott, Wilkins, Fuller & Langridge, 1967*a, b*; Arnott, Wilkins, Fuller, Venable & Langridge, 1967).

These crystalline polynucleotides do not occur as single crystals but as ordered arrays of microcrystals in fibres. The number of X-ray diffracted intensities obtainable from such systems is characteristically not more than a few hundreds, and few represent periodicities less than 2.5 Å. To define atomic positions in these molecules it is therefore necessary to supplement the diffraction data with additional stereochemical information, mainly in the form of the expected bond

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