CRYSTALLOGRAPHIC DATA FOR SOME AMINO ACIDS, DIPEPTIDES AND RELATED COMPOUNDS*

K. K. CHACKO, R. CHANDRASEKHARAN, MRS. GODAVARI CHANDRASEKHARAN,**
A. R. KALYANARAMAN, M. MALLIKARJUNAN, R. RAMACHANDRA AYYAR,
M. N. SABESAN, T. SRIKRISHNAN, R. SRINIVASAN, E. SUBRAMANIAN,
S. THYAGARAJA RAO AND K. VENKATESAN

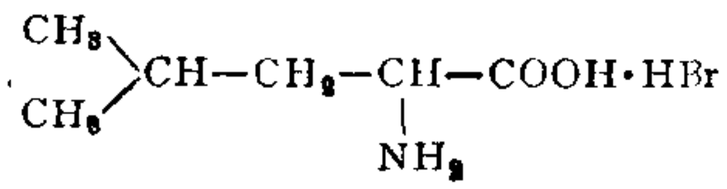
Centre of Advanced Study in Physics, University of Madras, Madras-25, India

WE present here preliminary crystallographic data for some amino acids, dipeptides and related compounds. The X-ray analysis of the structure of such compounds has been a long-term project at this Centre as part of a major programme on the structure of proteins and other related biomolecular substances. The data presented here pertain mainly to the determination of the unit cell dimensions and space group. The compounds reported here are: (1) L-leucine hydrobromide; (2) L-tryptophan hydrobromide; (3) DL-ornithine hydrobromide; (4) L-ornithine hydrochloride; (5) DL-histidine dihydrochloride; (6) DL-valine; (7) N-methyl-DL-leucyl glycine hydrobromide; (8) L-threonyl *p*-nitrobenzyl ester hydrobromide; (9) L-prolyl-L-phenylalanine O-methoxy hydrobromide; (10) L-prolyl-L-phenylalanine O-benzyl hydrobromide; (11) γ-L-glutamyl-L-methionine; (12) Chloroacetyl glycyl glycine; (13) Phenylenediamine dihydrochloride; (14) 1-amino cyclopentane carboxylic acid hydrobromide; (15) 1-amino cycloheptane carboxylic acid hydrobromide; and (16) 1-amino cyclo octane carboxylic acid hydrobromide. Of these compounds (1) to (6) are amino acid derivatives while (7) to (12) are dipeptide derivatives. The interest in compound (13) is that it has amino groups. Recently, much valuable information has been gleaned by a systematic study of the conformation of amino acids and other simple peptides (*e.g.*, Ramachandran *et al.*, 1966). It would also be of interest to obtain information about the conformation of related molecules containing amino and carboxylate groups under various other conditions and environments. It was for this reason the last three compounds (14) to (16) were also taken up which are cyclic compounds containing amino and carboxylate groups.

The structure in all essential details has been established for the compounds L-leucine hydrobromide, L-tryptophan hydrobromide, phenylenediamine dihydrochloride and L-threonyl-L-phenylalanine *p*-nitrobenzyl ester hydrobromide. The detailed reports of these will be published separately. The others are in various stages of analysis. A short description with regard to each compound is given below. Table I gives the crystallographic data. Unless otherwise stated, the determination of cell parameters was carried out mostly using Weissenberg and Precession photographs. The radiation used was CuK_α.

1. L-LEUCINE HYDROBROMIDE

The chemical formula of this compound is



Crystals were obtained by dissolving L-leucine in 30% hydrobromic acid and allowing the solution to evaporate at room temperature. The crystals were soft and hygroscopic and decomposed quickly on exposure to air. The crystal had to be sealed in Lindemann tube. The only systematic absences noticed were $h\ 0\ 0$ reflections, h odd absent; $0\ k\ 0$, k odd absent and $0\ 0\ l$, l odd absent. The space group was uniquely fixed as $P2_12_12_1$.

The structure was solved by the heavy atom method. The co-ordinates of the bromine atom were determined from the projection data along the *c*-and *a*-axes. The x co-ordinate of bromine turned out to be close to 0·25 which resulted in practically no contribution from the bromine atom to reflections of the type $h + k = 2n + 1$. Direct sign determining method was used to determine the signs of these reflections, where the signs determined for other reflections for bromine contribution were made use of. The structure was refined using three-dimensional data and the least square technique.

---

* Contribution No. 201 from the Centre of Advanced Study in Physics, University of Madras, Madras-25.

** Deceased.

TABLE I

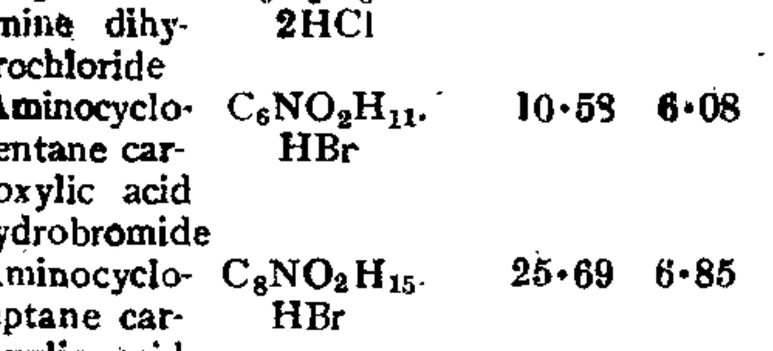
Crystalloghaphic data

Sl. No.	Name	Formula	a(Å)	b (Å)	c (Å)	α(°)	β(°)	γ(°)	V (Å^3)	ρ obs. gm./c.c.	Z	Molecular weight calculated	Molecular formula weight	Space group
1	L-Leucine hydrobromide	$C_6NO_2H_{13}$. HBr	7·29	24·51	5·54	..	..	..	989·9	$1·42_8$	4	210·5	212·0	$P2_12_12_1$
2	L-Tryptophan-hydrobromide	$C_{11}N_2O_2H_{12}$. HBr	14·56	5·44	7·57	..	100·50	..	591·9	$1·58_3$	2	282·0	285·0	$P2_1$
3	DL-Ornithine hydrobromide	$C_5N_2O_2H_{12}$. HCBr	9·39	7·90	11·66	..	109·83	..	813·6	$1·74_7$	4	213·9	213·0	$P2_1/c$
4	L-Ornithine hydrochloride	$C_5N_2O_2H_{12}$. HCl	5·02	8·03	10·05	..	97·05	..	402·1	$1·35_0$	2	163·5	168·0	$P2_1$
5	DL-Histidine dihydrochloride	$C_6N_3O_2H_9$. 2HCl	8·38	15·42	8·81	..	113·55	..	1043·5	$1·48_7$	4	230·7	228·0	$P2_1/c$
6	DL-Valine	$C_5NO_2H_{11}$	5·20	22·12	5·41	..	109·03	..	587·8	$1·32_0$	4	116·2	117·0	$P2_1/c$
7	N-Methyl-leucyl glycine-hydrobromide	$C_9N_2O_3H_{18}$. HBr	17·21	17·80	8·41	..	..	..	2576·3	$1·47_2$	8	283·3	283·0	$Pbca$
8	L-Threonyl-L-phenylalanine-*p*-nitrobenzyl ester hydrobromide	$C_{20}N_3O_6H_{25}$. HBr	8·93	45·75	5·05	..	..	..	2063·2	$1·60_9$	4	497·6	484·0*	$P2_12_12_1$
9	L-Prolyl-L-phenylalanine-O-methoxy hydrobromide	$C_{15}N_2O_3H_{20}$. HBr.	13·41	6·75	9·43	..	103·5	..	830·0	$1·41_0$	2	352·9	357·0	$P2_1$
10	L-Prolyl-L-phenylalanine-O-benzyl hydrobromide	$C_{20}N_2O_3H_{22}$. HBr	5·34	9·94	43·22	..	..	..	2294·1	$1·36_0$	4	470·3	419·0†	$P2_12_12_1$
11	γ-L-Glutamyl-L-methionine	$C_{10}N_2O_5H_{18}S$	5·09	9·83	26·56	..	..	..	1328·9	$1·42_0$	4	284·5	278·0	$P2_12_12_1$
12	Chloroacetyl glycylglycine	$C_6N_2O_4H_9Cl$	21·30	4·81	9·30	..	114·6	..	865·0	$1·57_0$	4	203·3	208·0	$P2_1/a$
13	Phenylenediamine dihydrochloride	$C_6N_2H_8$. 2HCl	8·75	5·87	4·34	99·78	95·57	111·17	201·4	$1·50_8$	1	181·4	181·0	$P\bar{1}$
14	1-Aminocyclopentane carboxylic acid hydrobromide	$C_6NO_2H_{11}$. HBr	10·53	6·08	7·04	..	99·67	..	446·3	$1·62_0$	2	216·1	210·0	$P2_1$
15	1-Aminocycloheptane carboxylic acid hydrobromide	$C_8NO_2H_{15}$. HBr	25·69	6·85	6·61	..	..	..	1162·3	$1·47_4$	4	256·2	238·0‡	$P2_12_12_1$
16	1-Aminocyclooctane carboxylic acid hydrobromide	$C_9NO_2H_{17}$. HBr	26·40	7·09	6·16	..	..	..	1153·0	$1·47_3$	4	253·8	252·0	$P2_12_12_1$

* See text ; † Indicates possibility of three water molecules in the asymmetric unit ; ‡ Indicates possibility of one water molecule in the asymmetric unit.

2. L-TRYPTOPHAN HYDROBROMIDE

The chemical formula of this is

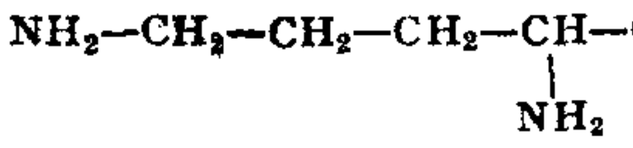


Crystals were prepared by dissolving L-tryptophan in hydrobromic acid and slowly evaporating the solution. The only systematic absences noticed were 0 k 0 reflections, k odd absent. The space group was thus fixed uniquely as $P2_1$. The structure was determined by the usual heavy atom method, first locating the heavy atom using projection data. Refinement

of the structure using three-dimensional data is in progress.

3. DL-ORNITHINE HYDROBROMIDE

The chemical formula of this compound is



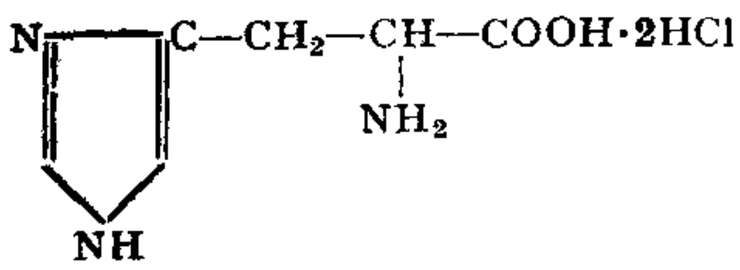
Crystals were obtained, as colourless needles, from aqueous solution by slow evaporation. The systematic absences were h 0 l reflections with l odd absent and 0 k 0 reflections k odd absent. Thus, the space group was uniquely fixed to be $P2_1/c$. The structure has been determined in the c projection and the three-dimensional work is in progress.

4. L-ORNITHINE HYDROCHLORIDE

The amino acid here is the same as in the previous one excepting that this is a hydrochloride derivative and an optically active form. Crystals were obtained from aqueous solution at room temperature. The crystal was found to belong to the monoclinic system with the needle as the non-unique axis (a). The only systematic absences were 0 k 0, k odd absent. The space group was thus fixed to be $P2_1$.

5. DL-HISTIDINE DIHYDROCHLORIDE

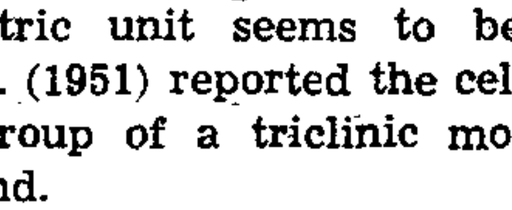
The chemical formula is



The crystals were obtained, with considerable difficulty by varying the conditions of crystallization. Small elongated crystals having bipyramidal shape were obtained. The system was found to be monoclinic. The systematic absences were 0 k 0, k odd absent and h 0 l, l odd absent. The space group was thus fixed to be $P2_1/c$.

6. DL-VALINE

The chemical formula is

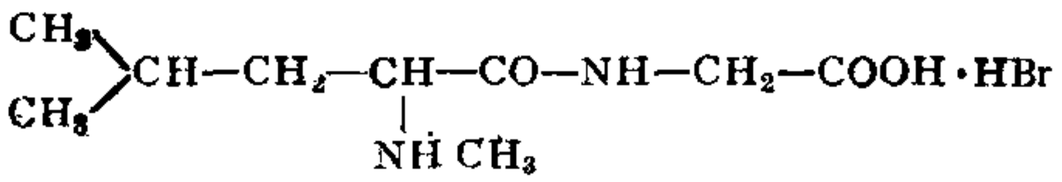


Crystals were picked from commercially available sample of DL-valine. The crystal was found to belong to the monoclinic system and the systematic absences were 0 k 0 reflections, k odd absent and h 0 l, l odd absent. These fixed the space group uniquely as $P2_1/c$.

Albrecht *et al.* (1943) reported the cell dimensions of this compound which are in essential agreement with our preliminary measurements. However, their assignment of the space group as $P2_1$ with two molecules in the asymmetric unit seems to be erroneous. Dawson *et al.* (1951) reported the cell dimensions and space group of a triclinic modification of this compound.

7. N-METHYL-DL-LEUCYL GLYCINE HYDROBROMIDE

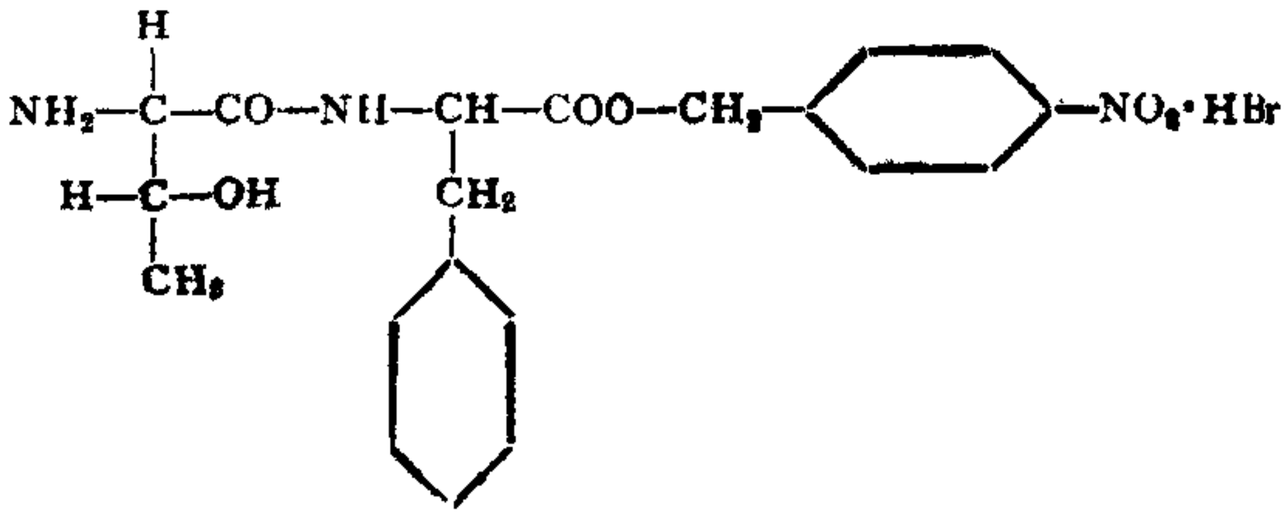
The chemical formula is



The crystal belongs to the orthorhombic system. The systematic absences were 0 k l, k odd absent, h 0 l, l odd absent, h k 0, h odd absent. The space group was thus fixed to be *Pbca*. The position of bromine atom has been determined from projections and the structure determination using three-dimensional data is in progress.

8. L-THREONYL-L-PHENYLALANINE *p*-NITROBENZYL ESTER HYDROBROMIDE

The chemical formula is



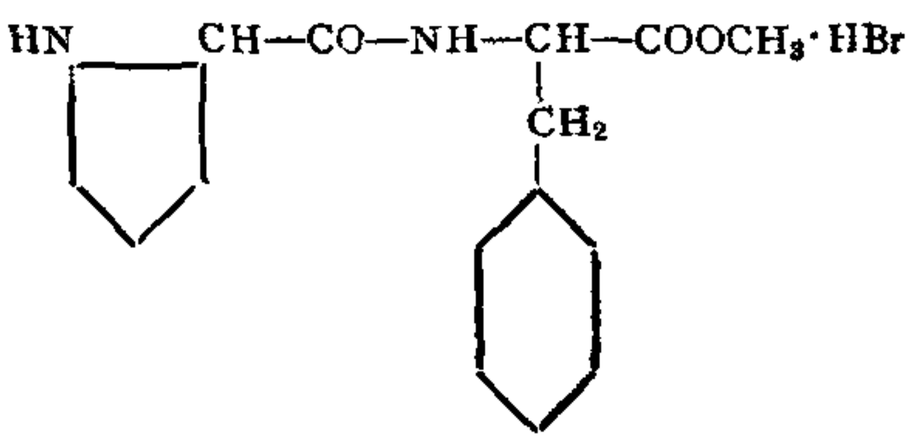
The crystals were obtained in the form of thin needles elongated along the c-axis by evaporating an aqueous solution at 45° C. The crystal belongs to the orthorhombic system.

From systematic absences the space group was fixed to be $P2_12_12_1$. The density, measured by flotation, was 1·60 gm./cm.^{3} Assuming four molecules in the unit cell the calculated molecular weight was found to be 497·6, the value corresponding with the chemical formula being 484.

The structure was determined *via* the heavy atom method and refinement by the Least-Squares method, using three-dimensional intensity data was carried out upto an R-value of 13·7%. The three-dimensional difference Fourier map calculated was almost featureless and did not provide any evidence for the existence of a water of crystallisation as may be suspected by molecular weight calculations.

9. L-Prolyl-L-Phenylalanine-O-Methoxy Hydrobromide

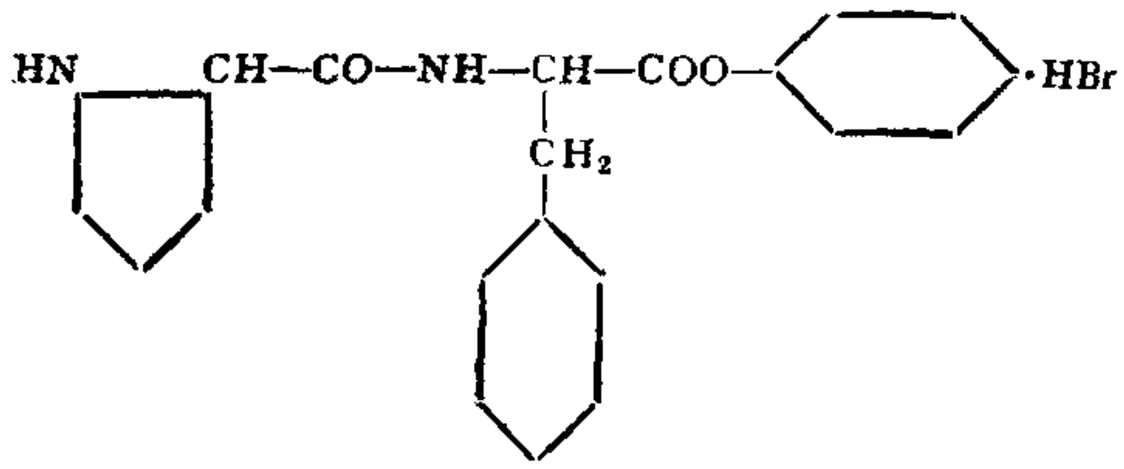
The chemical formula is



The crystals were platelike in shape elongated along the b-axis; c* was normal to the principal faces of the plates. The crystal belongs to the monoclinic system. The only systematic absences were 0 k 0, k odd absent, which indicated that the space group is $P2_1$. The density, measured by flotation, was 1·41 gm./c.c. and the calculated density was 1·43 gm./c.c., assuming two molecules per unit cell. The position of bromine atom has been determined in the b-axis Patterson projection. The determination of the structure in projection down the b-axis is in progress.

10. L-Prolyl-L-Phenylalanine-O-Benzyl Hydrobromide

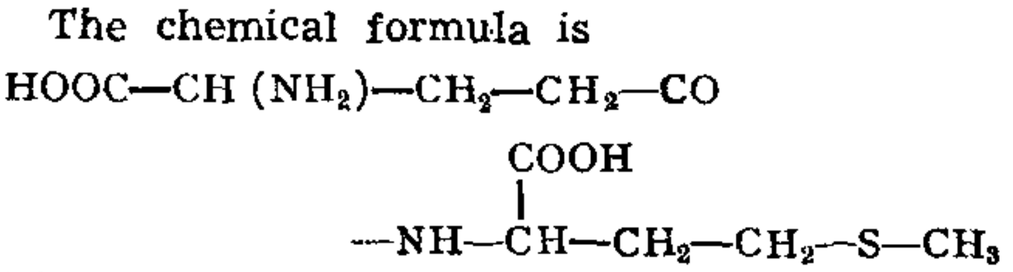
The chemical formula is



The crystals were in the form of thin needles elongated along the *a*-axis. The crystal belongs to the orthorhombic system. From systematic absences, the space group was fixed to be $P2_12_12_1$. The density measured by flotation was 1·36 gm./cm.^{3} With four molecules in the unit cell, the calculated molecular weight of the crystallographic asymmetric unit was 470·3. The value corresponding with the chemical formula was 419. The difference suggests presence of three water molecules in the asymmetric unit. The crystals were hygroscopic and were sealed in Lindemann capillary tubes during photography.

11. γ-L-Glutamyl-L-Methionine

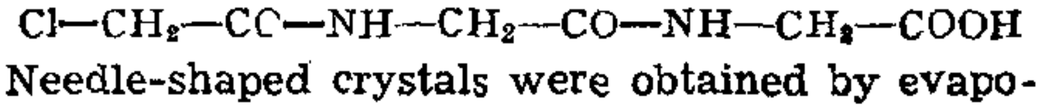
The chemical formula is



The compound isolated from the bulbs of onion (*Allium cepa*) was kindly sent by Professor A. I. Virtanen. Colourless thin plate-like crystals were recrystallized from acetone-water solution. The crystals were found to be orthorhombic and from systematic absences the space group was fixed to be $P2_12_12_1$, with four molecules in the unit cell.

12. Chloroacetyl Glycyl Glycine

The chemical formula is



Needle-shaped crystals were obtained by evaporating an aqueous solution of the commercially available sample at 40° C. They were elongated along the *b*-axis. Rotation and Weissenberg photographs showed the system to be monoclinic. The density measured by the method of flotation was 1·57 gm./c.c. The systematic absences were 0 k 0 reflections, k odd and h 0 l, l odd absent. Thus, the space group was fixed to be $P2_1/c$.

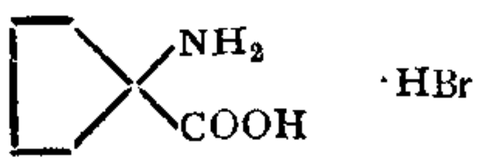
13. PHENYLENEDIAMINE DIHYDROCHLORIDE

The chemical formula is

Good crystals were picked from commercially available pure sample. The crystal had to be sealed in Lindemann tube since it decomposed on exposure to air. The crystal was found to belong to the triclinic system, with one molecule per unit cell. Since the molecule has a centre of symmetry the space group was assumed to be $P\bar{1}$ with half the molecule constituting the asymmetric unit. The structure was determined in projection and refinement using three-dimensional data is in progress.

14. 1-AMINO-CYCLOPENTANE CARBOXYLIC ACID HYDROBROMIDE

The chemical formula is

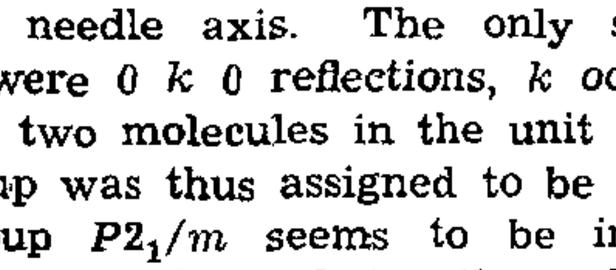


Samples of this and the heptane (15) and octane (16) derivatives described below were kindly supplied by Dr. R. Zand of the University of Michigan. The crystals were found to be needle-shaped.

The system was found to be monoclinic with b as the needle axis. The only systematic absences were $0\ k\ 0$ reflections, k odd absent. There are two molecules in the unit cell. The space group was thus assigned to be $P2_1$. The space group $P2_1/m$ seems to be improbable since this would demand that the molecule and the bromine atom should lie on symmetry plane which is too severe a restriction as seen from the nature of the formula. In fact, the structure in one projection namely along b-axis has been established, by first locating the bromine atom from the Patterson function.

15. 1-AMINO-CYCLOPENTANE CARBOXYLIC ACID HYDROBROMIDE

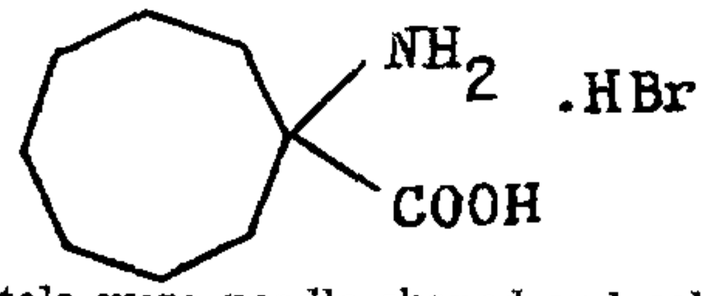
The chemical formula is



The crystals were thick and flaky in shape. The system was found to be orthorhombic. Systematic absences were $h\ 0\ 0$ reflections, h odd absent; $0\ k\ 0$, k odd absent and $0\ 0\ l$, l odd absent. The space group was fixed to be $P2_12_12_1$. The calculated value of molecular weight assuming $Z=4$ gave 256 while the expected formula weight was 238. The difference thus suggested presence of one water molecule per asymmetric unit.

16. 1-AMINO-CYCLOOCTANE CARBOXYLIC ACID HYDROBROMIDE

The chemical formula is



The crystals were needle-shaped and colourless. Systematic absences were $h\ 0\ 0$ reflections, h odd absent; $0\ k\ 0$, k odd absent and $0\ 0\ l$, l odd absent. The space group was fixed to be $P2_12_12_1$, with four formula units in the unit cell.

It may be noticed in particular that the cycloheptane and cyclooctane derivatives have unit cell dimensions which are rather close. The space groups also suggest that they are possibly isomorphous. The volume of the unit cell of the heptane derivative is 1162 Å^3, while that of the octane derivative is 1157 Å^3. The one CH_2 group that is less in the heptane compared to the octane one, appears to have been compensated by the presence of one water molecule in the former. These require full confirmation by actual structure analysis.

A better assessment of the possible isomorphism between the two crystals would be to use the statistical methods (Ramachandran *et al.*, 1963; Srinivasan *et al.*, 1963). These are to be made after intensity data become available and will be reported in due course.

We wish to record our thanks to Professor G. N. Ramachandran for his interest in the work. Thanks are due to Professor S. Akabori and Dr. S. Sakakibara of the Institute of Protein Research, Osaka, for supplying samples of compound (8); to Professor E. Havinga, Rijiksuniversiteit, Leiden, for sending us compounds (9) and (10); to Professor A. I. Virtenan, Biochemical Institute, Helsinki, for giving samples of (11) and to Dr. R. Zand of the Biophysics Research Division, University of Michigan, for kindly supplying samples of the compounds (14), (15) and (16).

1. Albrecht, G., Schnakenberg, G. W., Dunn, M.S., and McCullough, J. D., *J. Phys. Chem.*, 1943, **47**, 24
2. Dawson, G. and Mathieson, A. McL., *Acta Cryst.*, 1951, **4**, 475.
3. Ramachandran, G. N., Mazumdar, S. K., Venkatesan, K. and Lakshminarayanan, A. V., *J. Mol. Biol.*, 1966, **15**, 232.
4. —, Srinivasan, R. and Raghupathy Sarma, V., *Acta Cryst.*, 1963, **16**, 662.
5. Srinivasan, R. Raghupathy Sarma, V. and Ramachandran, G. N., In *Crystallography and Crystal Perfection*, Ed. G. N. Ramachandran, Academic Press, London, 1963, p. 85.