

# Crystal Structures of Diketopiperazines Containing $\alpha$ -Aminoisobutyric Acid: Cyclo(Aib-Aib) and Cyclo(Aib-L-Ile)

K. SUGUNA, S. RAMAKUMAR, N. SHAMALA,  
B. V. VENKATARAM PRASAD, and P. BALARAM, *Molecular  
Biophysics Unit and Department of Physics, Indian Institute of  
Science, Bangalore 560 012, India*

## Synopsis

The crystal and molecular structures of two  $\alpha$ -aminoisobutyric acid (Aib)-containing diketopiperazines, cyclo(Aib-Aib) **1** and cyclo(Aib-L-Ile) **2**, are reported. Cyclo(Aib-Aib) crystallizes in the space group  $P\bar{1}$  with  $a = 5.649(3)$ ,  $b = 5.865(2)$ ,  $c = 8.363(1)$ ,  $\alpha = 69.89(6)$ ,  $\beta = 113.04(8)$ ,  $\gamma = 116.0(3)$ , and  $Z = 1$ , while **2** occurs in the space group  $P2_12_12_1$  with  $a = 6.177(1)$ ,  $b = 10.791(1)$ ,  $c = 16.676(1)$ , and  $Z = 4$ . The structures of **1** and **2** have been refined to final  $R$  factors of 0.085 and 0.086, respectively. In both structures the diketopiperazine ring shows small but significant deviation from planarity. A very flat chair conformation is adopted by **1**, in which the  $C^\alpha$  atoms are displaced by 0.07 Å on each side of the mean plane, passing through the other four atoms of the ring. Cyclo(Aib-Ile) favors a slight boat conformation, with Aib  $C^\alpha$  and Ile  $C^\alpha$  atoms displaced by 0.11 and 0.05 Å on the same side of the mean plane formed by the other ring atoms. Structural features in these two molecules are compared with other related diketopiperazines.

## INTRODUCTION

Cyclic peptides are useful systems for investigation of the conformational characteristics of peptides.<sup>1</sup> Diketopiperazines (2,5-piperazinediones) are the simplest models for the study of *cis*-peptide geometries and for examining the influence of side chains on ring conformations.<sup>2,3</sup> The diketopiperazines investigated so far have contained the usual L- and D-amino acids and N-alkylated residues such as sarcosine,<sup>4,5</sup> proline,<sup>6-9</sup> or N-methyl amino acids.<sup>10,11</sup> Interest has largely been centered on the conformations of the six-membered ring and on side-chain backbone interactions.<sup>2,3</sup>  $\alpha$ -Alkylation has been shown to restrict the conformational freedom of acyclic peptides considerably.<sup>12</sup> This phenomenon has been exemplified in a number of experimental investigations of  $\alpha$ -aminoisobutyric acid-containing peptides.<sup>13</sup> It is therefore useful to examine the influence of  $\alpha$ -alkylation on cyclic peptide geometries. The only crystal structure available so far of an Aib-containing cyclic peptide is the cyclic tetrapeptide, dihydrochlamydocin.<sup>14</sup> In this report, we describe the crystal structures of the diketopiperazines cyclo(Aib-Aib) and cyclo(Aib-L-Ile).

TABLE I  
Crystal Data

Variable	c(Aib-Aib)	c(Aib-Ile)
Molecular formula	$C_8N_2O_2H_{14}$	$C_{10}N_2O_2H_{18}$
Molecular weight (amu)	170	198
Crystal system	Triclinic	Orthorhombic
Space group	$P\bar{1}$	$P2_12_12_1$
Z (molecules/unit cell)	1	4
a (Å)	5.649 (3)	6.177 (1)
b (Å)	5.865 (2)	10.791 (1)
c (Å)	8.363 (1)	16.676 (1)
$\alpha$ (°)	69.890 (6)	90
$\beta$ (°)	113.040 (8)	90
$\gamma$ (°)	116.010 (3)	90
Volume (Å <sup>3</sup> )	224.080	1111.56
Density (g/cm <sup>3</sup> )	1.259	1.183

## EXPERIMENTAL

Cyclo(Aib-Aib) **1** and cyclo(Aib-Ile) **2** were synthesized as described earlier.<sup>15</sup> Single crystals of both peptides were obtained by slow evaporation of saturated methanol solutions. Crystal data for the peptides are summarized in Table I. Intensity data were collected on a CAD-4 diffractometer, using  $\omega$ - $2\theta$  scan and Ni-filtered  $CuK_{\alpha}$  radiation. We used 726 observed reflections, with  $I \geq 3\sigma(I)$ , in the structure determination of **1**, while 1022 reflections were used for **2**. Intensities were corrected for Lorentz and polarization factors, but not for absorption.

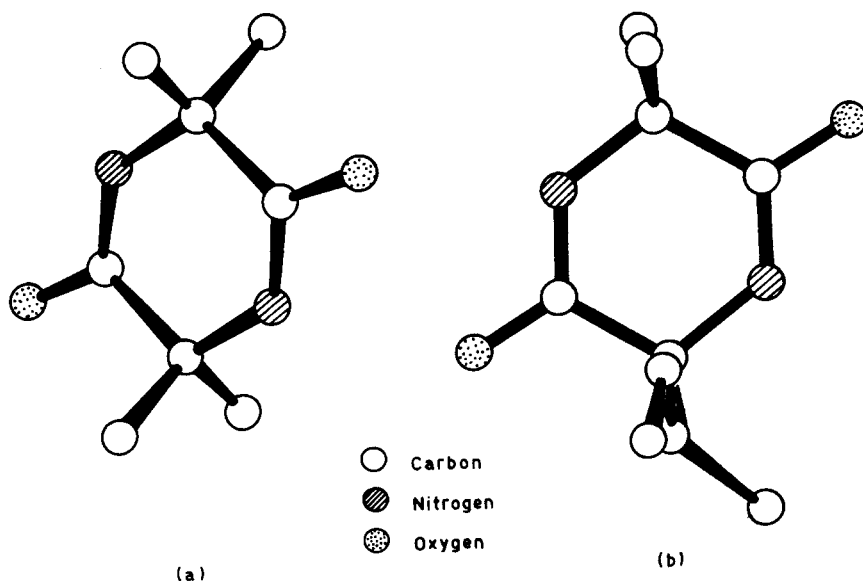
Fig. 1. Molecular structure of (a) cyclo(Aib-Aib) **1** and (b) cyclo(Aib-Ile) **2**.

TABLE II  
Final Atomic Coordinates and Isotropic Equivalent of Anisotropic Thermal Parameters for the Nonhydrogen Atoms of c(Aib-Aib) and c(Aib-Ile)

Atom	c(Aib-Aib)				c(Aib-Ile)				$B_{eq}$ ( $\text{\AA}^2$ )
	X	Y	Z	$B_{eq}$ ( $\text{\AA}^2$ )	X	Y	Z	$B_{eq}$ ( $\text{\AA}^2$ )	
N <sub>1</sub>	-0.1616 (6)	-0.2418 (5)	0.0583 (4)	2.75	0.2220 (9)	-0.0978 (6)	-0.0760 (3)	2.72	
C <sub>1</sub> <sup>a</sup>	0.1086 (7)	-0.1401 (5)	0.1856 (4)	2.26	0.4011 (10)	-0.0872 (7)	-0.1334 (4)	2.43	
C <sub>1</sub> <sup>b</sup>	0.2613 (10)	-0.3225 (7)	0.2392 (8)	3.89	0.3824 (12)	-0.1955 (7)	-0.1915 (4)	3.09	
C <sub>1</sub> <sup>c</sup>	0.0558 (10)	-0.1187 (8)	0.3451 (6)	4.51	0.3854 (14)	0.0367 (8)	-0.1785 (5)	3.95	
C <sub>1</sub>	0.2740 (7)	0.1302 (5)	0.1026 (4)	2.44	0.6195 (10)	-0.0901 (7)	-0.0911 (4)	2.58	
O <sub>1</sub>	0.5091 (5)	0.2365 (5)	0.1932 (3)	3.58	0.7852 (7)	-0.0998 (6)	-0.1325 (3)	3.52	
N <sub>2</sub>	—	—	—	—	0.6284 (8)	-0.0863 (6)	-0.0108 (3)	2.42	
C <sub>2</sub> <sup>a</sup>	—	—	—	—	0.4466 (10)	-0.0819 (7)	0.0451 (4)	2.61	
C <sub>2</sub> <sup>b</sup>	—	—	—	—	0.4529 (11)	0.0323 (7)	0.1007 (4)	2.79	
C <sub>2</sub> <sup>c</sup>	—	—	—	—	0.6402 (14)	0.0248 (9)	0.1569 (5)	4.88	
C <sub>2</sub> <sup>d</sup>	—	—	—	—	0.4400 (15)	0.1501 (7)	0.0514 (5)	3.78	
C <sub>2</sub> <sup>e</sup>	—	—	—	—	0.4029 (19)	0.2627 (10)	0.1039 (6)	5.61	
C <sub>2</sub>	—	—	—	—	0.2311 (10)	-0.0925 (8)	0.0021 (4)	2.60	
O <sub>2</sub>	—	—	—	—	0.0657 (8)	-0.0992 (6)	0.0461 (3)	3.66	

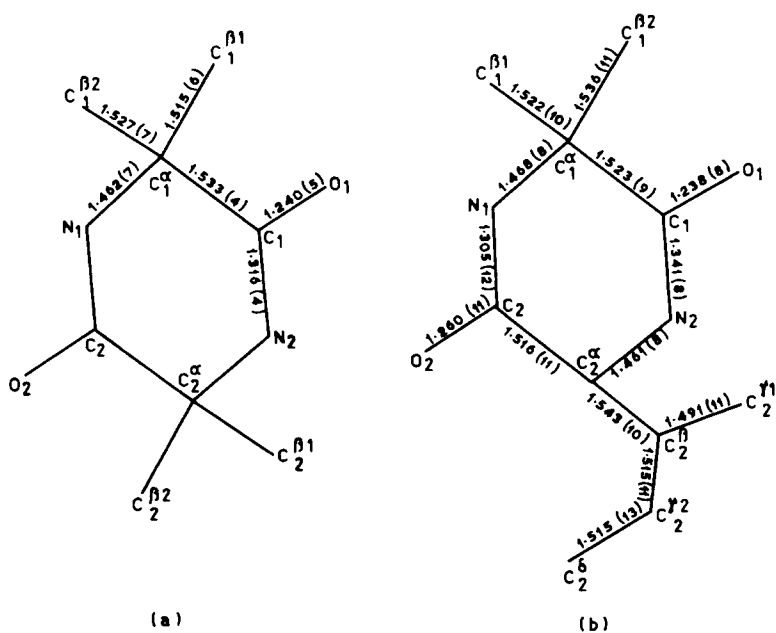


Fig. 2. Bond lengths ( $\text{\AA}$ ) in (a) cyclo(Aib-Aib) 1 and (b) cyclo(Aib-Ile) 2. ESDs are given in parentheses.

## Structure Determination and Refinement

### *Cyclo(Aib-Aib) 1*

The structure was solved in the space group  $P\bar{1}$  by direct methods using MULTAN.<sup>16</sup> A statistically weighted tangent formula<sup>17</sup> was used to calculate the phases of 191 reflections with  $E > 1.203$ . The  $E$  map, calculated with the phases corresponding to the set with the highest figure of merit, revealed 6 out of 12 atoms. The remaining 6 atoms were obtained by the Karle recycling process.<sup>18</sup> Using standard procedures, the structure refined to an  $R$  index of 0.085.

### *Cyclo(Aib-Ile) 2*

The structure was solved by the symbolic addition procedure, as applied to noncentrosymmetric space groups<sup>19,20</sup> and refined to a final  $R$  index of 0.086, using block diagonal least-squares procedures. The scattering factors used in the structure determination of 1 and 2 were those of Cromer and Waber<sup>21</sup> for nonhydrogen atoms and of Stewart et al.<sup>22</sup> for hydrogen atoms. The coordinates of the nonhydrogen atoms, along with the isotropic equivalent of anisotropic temperature factors<sup>23</sup> for both peptides, are listed in Table II. A listing of the observed and calculated structure factors, anisotropic thermal parameters for nonhydrogen atoms, and positional parameters of hydrogen atoms is available on request.

TABLE III  
Ring Bond Angles and Torsional Angles in Related Diketopiperazines

Diketopiperazine	Bond Angle (deg)			Conformational Angle (deg)			Conformational State	Ref.
	$\tau_a$	$\tau_N$	$\tau_C$	$\phi$	$\psi$	$\omega$		
Gly-Gly	115.1	126.0	118.9	1	-1	1	Planar	26
Sar-Sar	117.0	124.6	118.1	-1	1	-1	Chair	4
L-Ala-L-Ala	110.5	126.2	116.9	-7	7	-7	Boat	27
D-Ala-L-Ala	112.0	125.9	116.8	7	-7	7	Chair	27
Aib-Aib	113.4	127.9	118.6	-32	21	8	Chair	Present study
Aib-Ile	110.9	130.1	118.7	-25	27	1	Boat	Present study
	111.3	127.9	119.9	3	-3	3	Chair	Present study
	111.7	127.4	120.9	-3	3	-3	Boat	Present study
				-6	5	6		
				6	-5	-6		
				9.8	-8.0	-4.2		
				4.6	-3.4	1.4		

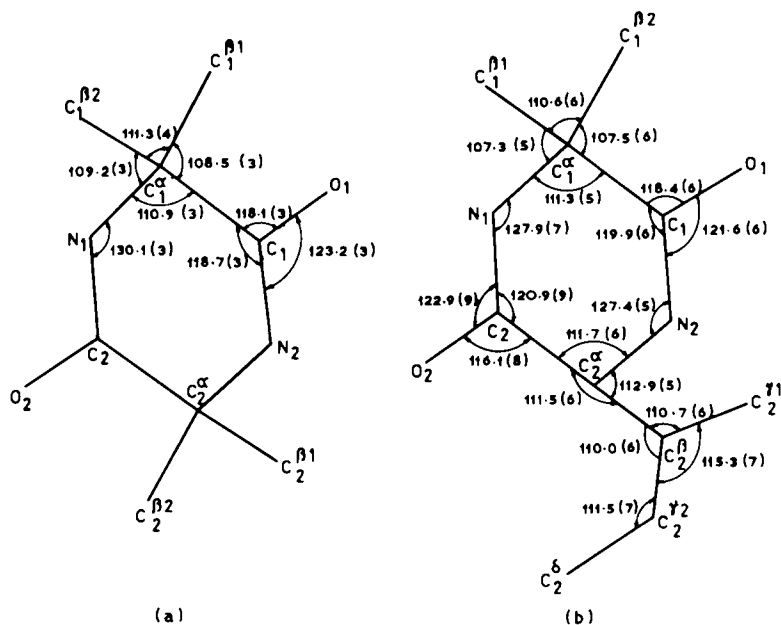


Fig. 3. Bond angles (Å) in (a) cyclo(Aib-Aib) 1 and (b) cyclo(Aib-Ile) 2. ESDs are given in parentheses.

## RESULTS AND DISCUSSION

The perspective view of the molecules 1 and 2 in the crystalline state are shown in Fig. 1. The bond lengths, bond angles, and ring torsional angles (defined according to the IUPAC-IUB Convention on Biochemical Nomenclature<sup>24</sup>) in 1 and 2 are summarized in Figs. 2–4, respectively. The bond lengths and bond angles are largely unexceptional and compare well with values reported for other diketopiperazines. There are some differences in the internal bond angles of the six-membered piperazinedione ring. The bond angles N—C<sup>α</sup>—C ( $\tau_{\alpha}$ ), C—N—C<sup>α</sup> ( $\tau_N$ ), and C<sup>α</sup>—C—N ( $\tau_C$ ) in the structures 1 and 2 are compared with relevant diketopiperazine structures in Table III. Alkylation at C<sup>α</sup> results in lowering  $\tau_{\alpha}$ , a feature seen for the Ala, Ile, and Aib residues. An interesting observation in 1 is the rather large value of  $\tau_N$ . The various bond angles about the Aib C<sup>α</sup> atom are very similar to the average values reported from an analysis of crystal structure data for acyclic Aib peptides.<sup>25</sup>

The ring torsional angles in 1 and 2 are compared with the related structures in Table III. In 1 the diketopiperazine ring is slightly distorted from the almost perfectly planar conformation found in the parent peptide cyclo(Gly-Gly).<sup>26</sup> The two Aib C<sup>α</sup> atoms are displaced by 0.07 Å on either side of the mean plane described by the other four atoms. The conformation may thus be viewed as a rather flat chair. In 2 the Aib C<sup>α</sup> and Ile C<sup>α</sup> are displaced by 0.11 and 0.05 Å on the same side of the mean plane formed by the other ring atoms, indicating a tendency towards a boat conformation. Diketopiperazines consisting of identical achiral residues

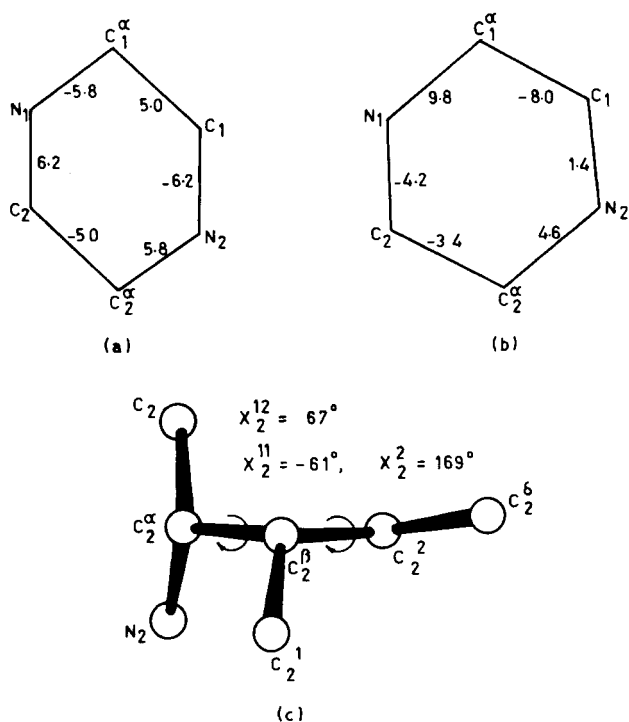


Fig. 4. Ring and side-chain torsional angles (deg) in (a) cyclo(Aib-Aib) 1 ring, (b) cyclo(Aib-Ile) 2 ring, and (c) Ile side chain.

or asymmetric residues of opposite chirality possess a center of symmetry. In such a case, only the planar and flat-chair conformations are generally observed. An exception, perhaps, is the case of cyclo(L-Val-D-Val), which is reported to have adopted a boat conformation.<sup>10</sup> Both C-alkylation as in 1 and N-alkylation, as in cyclo(Sar-Sar),<sup>4</sup> result in a distortion of the ring

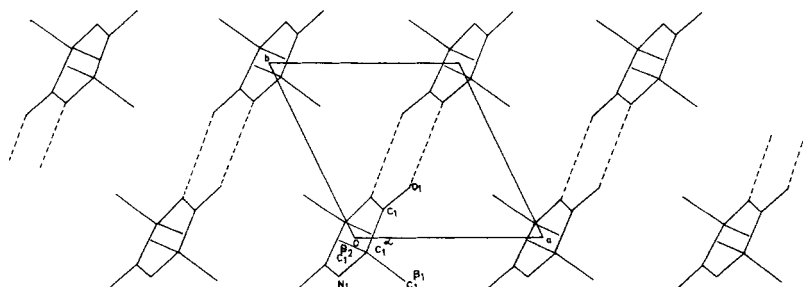


Fig. 5. Packing of cyclo(Aib-Aib) 1 molecules as viewed down the  $c^*$  axis. Dashed lines indicate intermolecular hydrogen bonds ( $N \cdots O$ , 2.89 Å;  $H-\tilde{N} \cdots O$ , 30.3°).

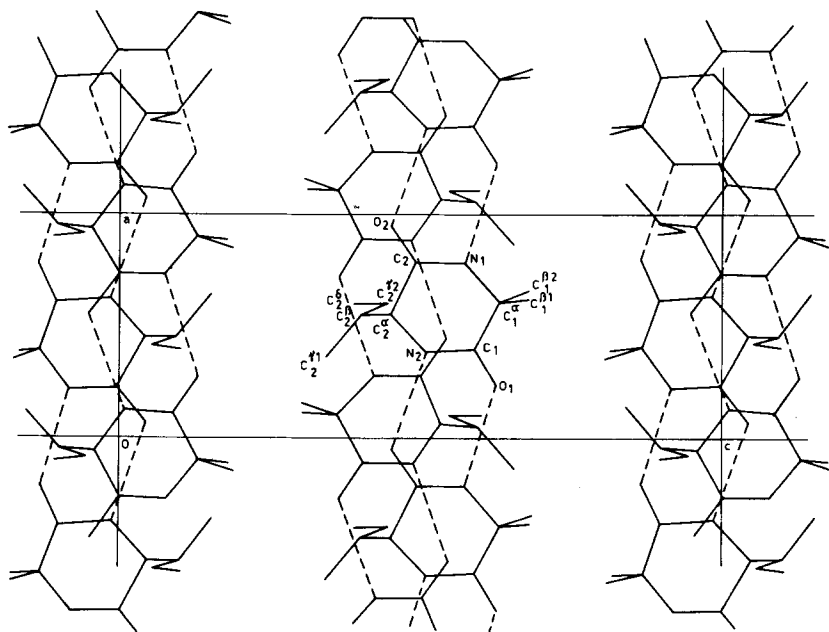


Fig. 6. Packing of cyclo(Aib-Ile) 2 molecules as viewed down the  $b$  axis. Dashed lines indicate intermolecular hydrogen bonds. The  $C_1O_1$  of the molecule at  $(x,y,z)$  and  $N_2H_2$  of the same molecule are hydrogen-bonded to  $N_1H_1$  ( $N\cdots O = 2.86 \text{ \AA}$ ,  $H-\tilde{N}\cdots O = 20.3^\circ$ ) and  $C_2O_2$  ( $N\cdots O = 2.87 \text{ \AA}$ ,  $H-\tilde{N}\cdots O = 12.3^\circ$ ), respectively, of the molecule at  $(1+x,y,z)$ .

to a flat chair. The structure of **1** also compares well with cyclo(D-Ala-L-Ala).<sup>27</sup> In **2**, where the ring is asymmetrically substituted, the boat form is favored, as in the case of cyclo(L-Ala-L-Ala)<sup>27</sup> and related structures like cyclo(L-Ser-L-Tyr),<sup>28</sup> and cyclo(Gly-L-Tyr),<sup>28</sup> and cyclo(Sar-L-Val).<sup>5</sup> However, the presence of Aib flattens the boat conformation considerably, as compared to cyclo(L-Ala-L-Ala). This is, presumably, to avoid the unfavorable 1-4 pseudoaxial interaction in a highly puckered conformation.

The conformation of the Ile side chain in **2** is illustrated in Fig. 4. Both substituents, at  $C^\beta$ , are *gauche* with respect to the  $C^\alpha-N$  bond. The conformation about  $C^\beta-C^\gamma$  is *trans* ( $\chi_2^2 = 169^\circ$ ). The side-chain atoms are extended away from the ring, with the ethyl group lying approximately perpendicular to the plane of the ring.

The intermolecular hydrogen-bonding pattern in the structures of **1** and **2** are similar (Figs. 5, 6) and correspond to a predicted mode of packing of diketopiperazines in the solid state.<sup>29</sup>

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