Acta Crystallographica Section E
**Structure Reports Online**
ISSN 1600-5368

# Tripeptide Z–Aib–Aib–Hyp–OMe

**Renate Geßmann,[a] Hans Brückner[b] and Michael Kokkinidis[a,c]***

[a]Institute of Molecular Biology and Biotechnology, Foundation of Research and Technology, PO Box 1527, 71110 Iraklion, Crete, Greece, [b]Institute of Nutritional Science, Department of Food Sciences, Interdisciplinary Research Center (IFZ), University of Giessen, Heinrich-Buff-Ring 26-32, 35392 Giessen, Germany, and [c]Department of Biology, University of Crete, Vassilika Vouton, 71409 Iraklion, Crete, Greece

Correspondence e-mail: kokkinid@crystal1.imbb.forth.gr

**Key indicators**

Single-crystal X-ray study
$T$ = 293 K
Mean $\sigma$(C–C) = 0.010 Å
Disorder in solvent or counterion
$R$ factor = 0.078
$wR$ factor = 0.211
Data-to-parameter ratio = 7.4

For details of how these key indicators were automatically derived from the article, see http://journals.iucr.org/e.

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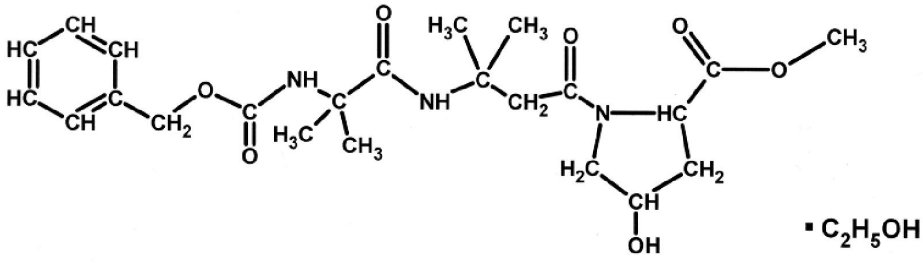
The structure of the synthetic protected tripeptide methyl 1-{*N*-[*N*-(benzyloxycarbonyl)-$\alpha$-aminoisobutyryl]-$\alpha$-aminoisobutyryl}-4-hydroxypyrrolidine-2-carboxylate (Z–Aib–Aib–Hyp–OMe) ethanol solvate, $C_{22}H_{31}N_3O_7.C_2H_6O$, was determined by X-ray crystallography. The peptide backbone adopts a conformation, which lies in the right-handed helical region for Aib1, in the left-handed helical region for Aib2 and in the semi-extended region for Hyp3.

Received 20 February 2001
Accepted 3 May 2001
Online 10 May 2001

Nomenclature: Aib is $\alpha$-aminoisobutyric acid, Hyp is L-hydroxyproline, (*trans*-4-hydroxy-L-proline), Z is benzyloxycarbonyl, OMe is methyl ester, and OBg is benzhydryl glycolamide ester.

## Comment

The tripeptide Aib–Aib–Hyp represents a segment of the naturally occurring peptaibol antibiotics [peptides containing $\alpha$-aminoisobutyric acid (Aib) and a C-terminal $\beta$-amino alcohol (Brückner & Graf, 1983; Benedetti *et al.*, 1982)] antiamoebins (Rinehart, 1983; Pandey, Meng *et al.*, 1977; Jaworski & Brückner, 2000) and emerimicins III/IV (Pandey, Cook & Rinehart, 1977; Brückner *et al.*, 1983). The conformational space available for Aib-residues comprises the right-handed $3_{10}$- and $\alpha$-helical region, as well as the left-handed $3_{10}$- and $\alpha$-helical region. Crystal structures of Antiamoebin I have been solved (Karle *et al.*, 1998; Snook *et al.*, 1998) and the segment Aib–Aib–Hyp adopts there $\varphi,\psi$-backbone torsion angles of −72/−31, −55/−46, −61/−20; −66/−35, −47/−48, −66/−16 and −68/−37, −48/−49, −63/−17°, *i.e.* all conformations lie in the right-handed helical region.




(I)

In the synthetic tripeptide, (I), the Aib residues adopt $\varphi,\psi$ values of −59.7, −44.7 (Aib1) and 58.1, 45.2° (Aib2); these correspond to the right- and left-handed helical regions, respectively. The hydroxyproline residue lies in the semi-extended region with $\varphi,\psi$ −67.1, 158.5. Overall similar backbone conformations have been observed for the tripeptides Z–Aib–Aib–Pro–OH and Z–Aib–Aib–Pro–OBg (Geßmann, 1999) for the Aib-residues, but not for proline, which lies in the other peptides in the right-handed helical region. The

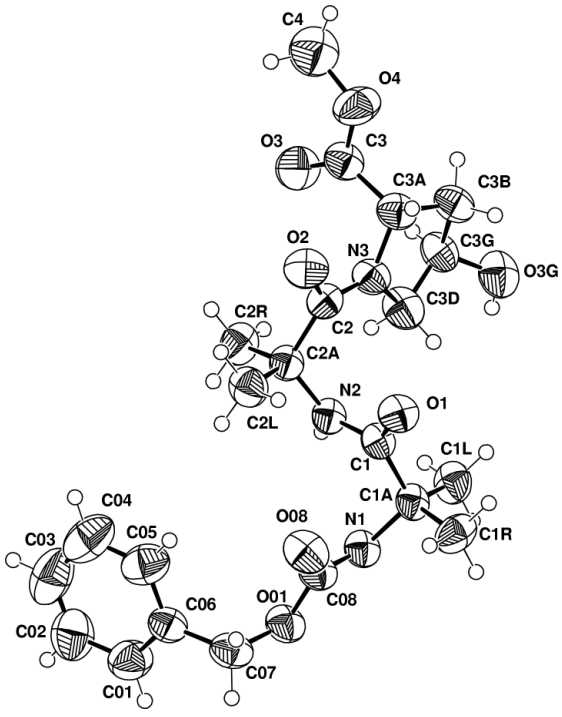



**Figure 1**
The molecular structure of (I) showing 50% probability displacement ellipsoids (Johnson, 1976; Davenport *et al.*, 2000).

valence geometry around the C$\alpha$ atoms is asymmetric for Aib residues. If one designates as C$i$L and C$i$R ($i$ is the identifier of the Aib residue) the atoms in Aib which occupy the same position as C$\beta$ and the $\alpha$H atom, respectively, in L-amino acids, then in right-handed helical conformations, the bond angles N$i$—C$i\alpha$—C$i$L and C$i$—C$i\alpha$—C$i$L are significantly smaller than the tetrahedral value (109.45°), and the angles N$i$—C$i\alpha$—C$i$R and C$i$—C$i\alpha$—C$i$R are larger (Table 1, Aib1). The opposite asymmetry is found for Aib2. The pyrrolidine ring of Pro adopts the C$_\gamma$-*endo* (Ashida & Kakudo, 1974) conformation with puckering parameters (Cremer & Pople, 1975) $Q$ = 0.318 Å and $\Phi$ = 96.19°. Each molecule is hydrogen bonded to its adjacent, disordered ethanol molecule. In the crystal, each molecule is hydrogen bonded by three hydrogen bonds to one other molecule, related by space-group symmetry; one of these hydrogen bonds is intermediated by the solvent O12 atom (Table 2). If one considers the direction donor–acceptor as an arrow and looks down the $c$ axis of the crystal, then each molecule is bound to a molecule turned 90° clockwise and shifted a quarter of the unit cell along [00$\bar{1}$]. Thus, infinitely long columns of molecules, each molecule bonded by three hydrogen bonds to one symmetry related molecule as donor ('downwards') and by three hydrogen bonds to another symmetry-related molecule as acceptor ('upwards') are formed along the $c$ axis (Figs. 2 and 3). Along the a and $b$ axes, parallel columns form apolar contacts involving mainly the two protecting groups and the apolar end of the solvent molecule.

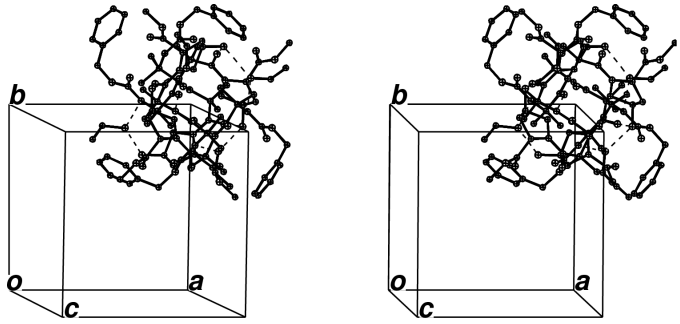



**Figure 2**
Crystal packing of Z–Aib–Aib–Hyp–OMe viewed approximately down the $c$ axis. The four space-group symmetry-related molecules are shown. Intermolecular hydrogen bonds are indicated by broken lines; H atoms have been omitted for clarity and the disordered solvent ethanol is shown in the main conformation (Johnson, 1976; Davenport *et al.*, 2000).

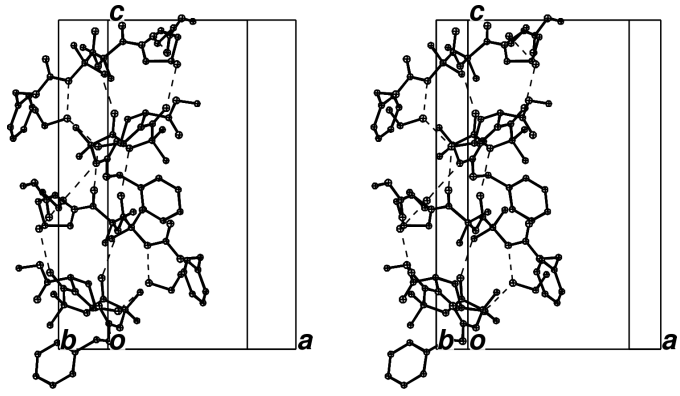



**Figure 3**
Crystal packing of Z–Aib–Aib–Hyp–OMe viewed approximately down the $b$ axis. The four space-group symmetry-related molecules are shown. Intermolecular hydrogen bonds are indicated by broken lines; H atoms have been omitted for clarity and the disordered solvent ethanol is shown in the main conformation (Johnson, 1976; Davenport *et al.*, 2000).

## Experimental

The tripeptide Z–Aib–Aib–Hyp–OMe was synthesized by heating the oxazolone of Z–Aib–Aib–OH [synthesized according to Jones *et al.* (1965)] with equimolar amounts (6.57 mmol) of H–L-Hyp–OMe·$x$HCl (from Bachem, Bubendorf, Switzerland) and *N*-methylmorpholine in acetonitrile for 16 h at 343 K. The mixture was evaporated to dryness. To the remaining residue chloroform (100 ml) was added, and the organic phase was washed three times each with 25 ml of 10% sodium carbonate, 5% sodium hydrogen sulfate and water. Then the organic phase was evaporated to dryness, ethyl acetate (50 ml) was added and the organic phase was dried over anhydrous sodium sulfate. Sodium sulfate was removed by filtration and the tripeptide was precipitated by addition of light petroleum. This approach does not cause racemization of Hyp, so the absolute configuration was assumed to be known. For analyses, a sample was crystallized from ethyl acetate. Yield 1.80 g (61%), m.p. 419 K; $[\alpha]_D^{20}$ −62.8 ($c$ = 1, MeOH); m/$z$ 450 $(M + H)^+$ elemental analysis for $C_{22}H_{31}N_3O_7$ (449.51), calculated: C 58.79, H 6.95, N 9.35%; found: C 58.88, H 7.01, N 9.24%. For X-ray analysis, the tripeptide was crystallized from a hot ethanol–water mixture (70:30).

*Crystal data*

$C_{22}H_{31}N_3O_7 \cdot C_2H_6O$
$M_r$ = 495.57
Tetragonal, $P4_1$
$a$ = 11.698 (2) Å
$c$ = 19.643 (15) Å
$V$ = 2688 (2) Å$^3$
$Z$ = 4
$D_x$ = 1.225 Mg m$^{-3}$
Cu $K\alpha$ radiation
Cell parameters from 25 reflections
$\theta$ = 12.7–23.9°
$\mu$ = 0.76 mm$^{-1}$
$T$ = 293 K
Rod, colourless
0.4 × 0.2 × 0.2 mm

*Data collection*

Enraf–Nonius CAD-4 diffractometer
$\omega$–$2\theta$ scans
Absorption correction: analytical (*Xtal*3.7 *ABSORB*; du Bouley & Hall, 2000)
$T_{min}$ = 0.80, $T_{max}$ = 0.88
5671 measured reflections
2622 independent reflections
2018 reflections with $I > 2\sigma(I)$
$R_{int}$ = 0.052
$\theta_{max}$ = 70.0°
$h = -14 \rightarrow 14$
$k = -14 \rightarrow 14$
$l = -21 \rightarrow 23$
5 standard reflections
frequency: 3600 min
intensity decay: 13.9%

*Refinement*

Refinement on $F^2$
$R[F^2 > 2\sigma(F^2)]$ = 0.078
$wR(F^2)$ = 0.211
$S$ = 1.08
2622 reflections
356 parameters
H-atom parameters constrained
$w = 1/[\sigma^2(F_o^2) + (0.1474P)^2]$ where $P = (F_o^2 + 2F_c^2)/3$
$(\Delta/\sigma)_{max} < 0.001$
$\Delta\rho_{max}$ = 0.43 e Å$^{-3}$
$\Delta\rho_{min}$ = −0.41 e Å$^{-3}$
Extinction correction: *SHELXL*97
Extinction coefficient: 0.0078 (13)
Absolute structure: assumed from synthesis

**Table 1**
Selected geometric parameters (Å, °).

| | | | |
|---|---|---|---|
| N1—C1*A*—C1*R* | 110.4 (4) | N2—C2*A*—C2*L* | 109.4 (4) |
| C1*R*—C1*A*—C1 | 111.1 (4) | C2*L*—C2*A*—C2 | 109.9 (5) |
| N1—C1*A*—C1*L* | 106.6 (4) | N2—C2*A*—C2*R* | 108.4 (4) |
| C1—C1*A*—C1*L* | 109.0 (4) | C2—C2*A*—C2*R* | 106.6 (4) |
| | | | |
| O01—C08—N1—C1*A* | −167.6 (4) | N3—C3*A*—C3—O4 | 158.5 (5) |
| C08—N1—C1*A*—C1 | −59.7 (6) | C3*A*—C3—O4—C4 | 173.8 (7) |
| N1—C1*A*—C1—N2 | −44.7 (6) | N3—C3*A*—C3*B*—C3*G* | 24.1 (6) |
| C1*A*—C1—N2—C2*A* | 172.2 (4) | C3*A*—C3*B*—C3*G*—C3*D* | −32.5 (6) |
| C1—N2—C2*A*—C2 | 58.1 (6) | C3*D*—N3—C3*A*—C3*B* | −6.5 (6) |
| N2—C2*A*—C2—N3 | 45.2 (7) | C3*B*—C3*G*—C3*D*—N3 | 28.0 (6) |
| C2*A*—C2—N3—C3*A* | 168.1 (5) | C3*A*—N3—C3*D*—C3*G* | −13.9 (7) |
| C2—N3—C3*A*—C3 | −67.1 (6) | | |

**Table 2**
Hydrogen-bonding geometry (Å, °).

| *D*—H···*A* | *D*—H | H···*A* | *D*···*A* | *D*—H···*A* |
|---|---|---|---|---|
| N1—H1···O12$^{i}$ | 0.86 | 2.27 | 3.003 (7) | 142 |
| N2—H2···O1$^{i}$ | 0.86 | 2.23 | 3.045 (5) | 157 |
| O3G—H30···O2$^{i}$ | 0.82 | 1.98 | 2.700 (6) | 147 |
| O12—H12···O3*G* | 0.92 | 1.96 | 2.874 (8) | 171 |

Symmetry code: (i) $y-1, 1-x, z-\frac{1}{4}$.

Data were collected from $\theta$ = 1–70°, 5005 unique data, including 2383 Friedel pairs with the Friedel opposite, if possible, measured at the negative $\theta$ position (from $\theta$ = 1 to −49°). All equivalent data were averaged, including the Friedel pairs. In an early stage of refinement with *Xtal*, a disorderd ethanol solvent molecule was observed. The two main conformations of the solvent were refined with distance restraints and without connectivity one to the other with *SHELXL* (Sheldrick & Schneider, 1997), together with the unrestrained tripeptide. H atoms were calculated, except the H atom associated with the O atom of ethanol, which was located from a difference Fourier synthesis. The riding model was used for all H atoms. Several displacement parameters of H atoms were fixed to $1.5U_{eq}$ of the parent atom due to physically unrealistically low or high refined values.

Data collection: *CAD*-4 *Software* (Enraf–Nonius, 1989); cell refinement: *CAD*-4 *Software*; data reduction: *Xtal*3.7 *DIFDAT ABSORB ADDREF* (du Bouley & Hall, 2000); program(s) used to solve structure: *Xtal*3.7 *CRISP* (du Bouley & Hall, 2000); program(s) used to refine structure: *SHELXL*97 (Sheldrick & Schneider, 1997); molecular graphics: *Xtal*3.7 *PIG* (du Bouley & Hall, 2000) and *Xtal*3.7 *ORTEP* (Davenport *et al.*, 2000); software used to prepare material for publication: *SHELXL*97, and *Xtal*3.7 *BONDLA* and *CIFIO* (du Bouley & Hall, 2000).

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