Crystal Structure of *t*-Butyloxycarbonyl-L-prolyl-L-hydroxyprolyl-glycine methyl ester (Boc-Pro-Hyp-Gly-OMe)

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The peptide Boc-Pro-Hyp-Gly-OMe (**I**) crystallizes in an orthorhombic system, and was characterized thus: $P2_12_12_1$, $a = 8.882(1)$, $b = 15.151(2)$, $c = 30.928(4)$ Å, $V = 4162.0(9)$ Å³, $Z = 4$. The final R value was 0.0454 for measured reflections. Two peptide molecules were found in the asymmetric unit with a somewhat twisted structure. These molecules were arranged antiparallel, creating a sheet-like structure. A down-pucker was observed for Pro residues, but the both up- and down-puckers were observed for the Hyp residues.

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Peptide (**I**), Boc-Pro-Hyp-Gly-OMe (Boc = *t*-butyloxycarbonyl; Hyp = (2*S*,4*R*)-hydroxyproline; OMe = methyl ester), was designed from triplets frequently found in collagens. We previously determined the structures of two related peptides: Boc-Phe-Hyp-Gly-OBzl (**II**) and Boc-Ala-Hyp-Gly-OBzl (**III**) (OBzl = benzyl ester).¹ In these structures, the conformations varied widely at the OBzl moiety. The bulky OBzl groups are presumed to affect the peptide conformation, and (**I**) was synthesized by a conventional liquid method using a small protect group of the C-terminal.

The structure was solved by direct methods, and non-H atoms were refined by a full-matrix least-squares method with anisotropic temperature factors. The positions of all H-atoms were geometrically calculated and refined isotropically with the riding model. The crystal and experimental data are given in Table 1. The two molecules (molecules A and B) with similar conformations are found in the asymmetric unit (Fig. 1). The molecules are slightly twisted, and the backbone torsion angles (Table 2) show that the structures of (**I**) still hold the characteristics of poly(Pro-Hyp-Gly).² However, the peptide molecules arrange to be antiparallel, and create a rather sheet-like structure in the crystal (Fig. 2). A hydrogen bond is formed between backbones: N30A(Gly)···O14B(Pro) = 2.857(3) Å and the hydroxyl group of Hyp (O22) participate in three hydrogen bonds: N30B(Gly)–H···O22A(Hyp) = 3.039(2) Å, O22A(Hyp)–H···O24A(Hyp) = 2.749(2) Å (the symmetry code of O24A: $1-x, y+1/2, 1/2-z$) and O22B(Hyp)–H···O14B(Pro) = 2.848(2) Å (the symmetry code of O14B: $-x, y+1/2, 1/2-z$). These interactions

stabilize this sheet-like structure.

The backbone torsion angles show a similarity between structures of molecules (**I**)-A and B, and molecular fitting is carried out. It seems that these structures are also similar to the previously reported structures of (**II**) and (**III**), and a total of six peptide molecules of three crystals are fitted to each other (Fig. 3). The entire dispersion among peptide backbones is notably low; RMS are 0.10–0.51 Å. The Gly residues are well matched among three structures (six peptides). This shows that the OBzl groups are less affected for the structures of peptide backbones. The conformational shifts are rather observed around Boc groups and following residues, but the C $_{\alpha}$ atoms of the first residues come close among three structures. The ring puckerings are shown along with the amplitude (Q_2) and phase (Φ_2);³ these values are listed in Table 3. The Pro/Hyp residues having Φ_2 around 280° and 80° are defined as up- and down-puckers, respectively. The (Q_2 , Φ_2) values of Pro indicate a

Fig. 1 Chemical diagram of the peptide (**I**).

Table 1 Crystal data and experimental details

Chemical formula: $2(C_{18}H_{29}N_3O_7)$	
Formula weight: 798.88	
$T = 240(2)$ K	
Crystal system: Orthorhombic	Space Groups: $P2_12_12_1$
$a = 8.882(1)$ Å	$V = 4162.0(9)$ Å ³
$b = 15.151(2)$ Å	$D_x = 1.275$ Mg m ⁻³
$c = 30.928(4)$ Å	$Z = 4$
Radiation: Mo K_{α} ($\lambda = 0.71073$ Å)	
$\mu(\text{Mo } K_{\alpha}) = 0.098$ mm ⁻¹	
$F(0\ 0\ 0) = 1712$	
No. of reflections collected = 45347 ($\theta_{\text{max}} = 27.1^\circ$)	
No. of reflections = 4162 [$I > 2\sigma(I)$]	
R indices [$I > 2\sigma(I)$]: $R1 = 0.0454$, $wR2 = 0.1093$	
R indices [all data]: $R1 = 0.0540$, $wR2 = 0.1128$	
Goodness-of-fit on $F^2 = 0.947$	
$(\Delta\sigma)_{\text{max}} = 0.001$	
$(\Delta\rho)_{\text{max}} = 0.270$ e Å ⁻³	$(\Delta\rho)_{\text{min}} = -0.177$ Å ⁻³
Measurement = Bruker SMART CCD	
Program system: WinGX	
Structure determination: direct methods (SHELXS-97)	
Refinement = full-matrix (SHELXL-97)	
CCDC deposition number: 751670	

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Table 2 Selected torsion angles ($^{\circ}$)

Bond	A	B
C5—N10—C10—C14 (Pro ϕ_1)	−68.9 (3)	−66.4 (3)
N10—C10—C11—C12 (Pro χ_1)	37.0 (3)	31.5 (3)
N10—C10—C14—N20 (Pro φ_1)	176.4 (2)	158.6 (2)
C10—C14—N20—C20 (Pro ω_1)	165.9 (2)	169.6 (2)
C14—N20—C20—C24 (Hyp ϕ_2)	−61.1 (3)	−76.5 (3)
N20—C20—C21—C22 (Hyp χ_2)	−28.0 (2)	31.5 (2)
N20—C20—C24—N30 (Hyp φ_2)	165.4 (2)	150.4 (2)
C20—C24—N30—C30 (Hyp ω_2)	173.1 (2)	169.4 (2)
C24—N30—C30—C31 (Gly ϕ_3)	−69.0 (3)	−77.5 (3)
N30—C30—C31—O32 (Pro φ_3)	178.8 (2)	175.9 (2)

Fig. 2 ORTEP plot with atom labeling (40% probability ellipsoids).

single puckering (down-pucker) in (I) and (II). However, two types of ring puckering of Hyp are clearly shown in Fig. 3, and the (Q_2 , Φ_2) values of Hyp are also classified into two groups. It is well-known that Hyp residues of poly(Pro-Hyp-Gly) favor down-pucker.⁴ It is suggested that both pucker of Hyp coexist in the short collagen peptide (I).

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Fig. 3 Stereoview of superimposition.

Table 3 Five-membered ring puckering amplitude (Q_2) and phase (Φ_2)

	(I)-A	(I)-B	(II)-A	(II)-B	(III)-A	(III)-B
Q_2 (Pro)	0.391(3)	0.363(3)	0.380	0.391	—	—
Φ_2 (Pro)	76.1(4)	85.5(4)	85	79	—	—
Q_2 (Hyp)	0.415(3)	0.328(3)	0.340	0.409	0.365	0.380
Φ_2 (Hyp)	282.2(3)	75.3(4)	81	286	282	267

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