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A Theoretical Study of Conformational Properties of *N*-Methyl Azapeptide Derivatives

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Abstract: The conformational properties of azapeptide derivatives, Ac-azaGly-NHMe (**1**), Ac-azaAla-NHMe (**2**), Ac-NMe-azaGly-NHMe (**3**), Ac-NMe-azaAla-NHMe (**4**), Ac-azaGly-NMe_2 (**5**), Ac-azaAla-NMe_2 (**6**), Ac-NMe-azaGly-NMe_2 (**7**), and Ac-NMe-azaAla-NMe_2 (**8**), were systematically examined by using ab initio MO and DFT methods. Structural perturbations in azapeptides resulting from cyclic substitution of a methyl group at three N-positions of an azaamino acid were studied on the basis of the structure of the simplest model azapeptide, **1**. Potential energy surfaces were generated at the HF/6-31G* level for **1**−**4** and at the HF/6-31G*//HF/3-21G level for **5**−**8** by rotating two key dihedral angles (ϕ, ψ) in increments of 30°. The backbone (ϕ, ψ) angles of the minima for **1**−**4** are observed at the $i+2$ position to form the βI(I′)-, βII(II′)-, βVI-turns or the polyproline II structure according to the orientation of the acetyl group and the positions of the *N*-methyl groups. Compounds **5**−**8** coupled to a secondary amine were found to preferentially adopt polyproline II, βI(III)-turn, or α-helical structure or even extended conformations depending on the orientation of the acetyl group and the positions of the *N*-methyl groups. Furthermore, *N*-methyl groups, depending on their positions, were found to affect the orientation of the amide group in the lowest energy conformations, the pyramidality of the N2 atom, and the bond length in azapeptide derivatives. These unique theoretical conformations of *N*-methyl azapeptide derivatives could be utilized in the definite design of secondary structure for peptides and proteins, and in the development of new drugs and molecular machines.

1. Introduction

The structure of proteins is controlled by varying the simplest elements of the secondary structure: helices, β-sheets, and β-turns. Among these structural elements of proteins, the β-turns are the most abundant regular elements connecting two β-strands.[1,2] Since the biological effects mediated by peptides greatly depend on their conformational properties, the introduction of geometrical constraints to the peptide structure could enhance the selectivity of biologically active peptides.[3]

Since bioactive peptides must adopt a specific conformation to bind to an acceptor molecule, the exploration of binding conformations is one of the most important processes involved in the effort to obtain potent and selective therapeutic agents. For this purpose, constrained peptidomimetics cause the resulting peptides to adopt distinct preferred conformations by removing the flexibility of the parent linear peptides.[3]

The identification of conformational preferences for peptidomimetics has become immensely important because of the need for predictable and controllable secondary structures in drug discovery and nanotechnology (i.e., peptide engineering, foldamers).[4−6] The ideal ϕ and ψ dihedral angles for regions of these secondary structures are shown in Table 1.[1,2,7] The various experimental and theoretical approaches to characterize

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Table 1. Ideal ϕ and ψ Dihedral Angles (in Degrees) for Residues of Defined Secondary Structure[a]

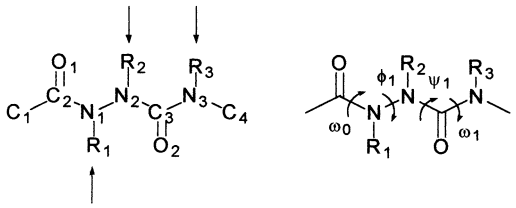
secondary structure	ϕ	ψ
right-handed α-helix	−57 (−65)[b]	−47 (−40)[b]
3_{10}-helix	−49 (−63)[b]	−26 (−17)[b]
γ-turn (2_7 ribbons)	−86	63
polyproline II	−78	149
polyglycine II	−80	150
β-strand	−180	180

β-turn	ϕ_{i+1}	ψ_{i+1}	ϕ_{i+2}	ψ_{i+2}
βI	−60	−30	−90	0
βI′	60	30	90	0
βII	−60	120	80	0
βII′	60	−120	−80	0
βIII	−60	−30	−60	−30
βIII′	60	30	60	30
βVIa[c]	−60	120	−90	0
βVIb′ [c]	−120	120	−60	150

[a] AIUPAC−IUB Commission on Biochemical Nomenclature [b] The backbone torsion angle, ϕ or ψ, in parentheses indicates the mean values (ref 7e). [c] The cis peptide bond is at the $i+1$ position (i.e., $\omega_{i+1} \approx 0°$).

the conformational properties of designed peptidomimetics in solution and crystalline states were performed by using NMR and X-ray spectroscopic methods and quantum mechanical calculations. These approaches have provided structural information regarding the accessible conformations of the modified amino acids in long peptides.[4−6]

Azaamino acids are formed by replacing an α-carbon of amino acids with a nitrogen atom without changing the side chains, as shown in Figure 1.[8−22] Since methods for their synthesis have been extensively investigated in solution and in the solid phase, many azaamino acids have been inserted into peptides and led to desirable bioactive moieties (i.e., luteinizing hormone-releasing hormone, angiotensin-coverting enzyme (ACE) inhibitor, and serine and cysteine proteases).[8−10] Despite extensive usage of azaamino acids in peptides, conformational studies have been relatively limited.[11−22] Aubry, Boussard, and Marraud have made major contributions to this area by carrying out crystallographic and spectroscopic studies of peptides containing azaamino acids.[11−16] Thormann and Hofmann intensively investigated the preferential conformations of various azapeptide models, including hydrazine, using several theoretical methods.[17] These experimental and theoretical studies demonstrated clearly that azaamino acids in peptides have characteristic structural properties.[11−22] The preferential backbone dihedral angles (ϕ, ψ) of azaamino acids in peptide structures are ($\pm 90° \pm 30°$, $0° \pm 30°$) and ($\pm 90° \pm 30°$, $\pm 180° \pm 30°$), which appear at the $i+2$ position of βI(II)-turn (δ_R) and polyproline II (β_P) structures, respectively. These imply that the incorporation of azaamino acid into a peptide would stabilize the β-turn or polyproline II conformation. We reported earlier that the azapeptides Ac-Aib-azaGly-NH_2, Boc-Phe-azaLeu-Ala-OMe, and Boc-Ala-Phe-azaLeu-Ala-OMe adopt the βI- and βII-turn conformations in solution, as observed using NMR spectroscopy.[19−21] The structures of azapeptides studied so far, in addition to these compounds, are shown in Table 2.[11−16,19−22] Since the X-ray and NMR structures available for azapeptide derivatives are considerably limited, it is significant to characterize the structures of various azapeptides coupled with primary and secondary amines at the C-terminus.



$R_1=R_2=R_3=H$ (**1**); $R_1=R_3=H$, $R_2=CH_3$ (**2**);
$R_1=CH_3$, $R_2=R_3=H$ (**3**); $R_1=R_2=CH_3$, $R_3=H$ (**4**);
$R_1=R_2=H$, $R_3=CH_3$ (**5**); $R_1=H$, $R_2=R_3=CH_3$ (**6**);
$R_1=R_3=CH_3$, $R_2=H$ (**7**); $R_1=R_2=R_3=CH_3$ (**8**)

Figure 1. Chemical structures and torsion angles for Ac-azaGly-NHMe (**1**), Ac-azaAla-NHMe (**2**), Ac-(NMe)azaGly-NHMe (**3**), Ac-(NMe)azaAla-NHMe (**4**), Ac-azaGly-NMe_2 (**5**), Ac-azaGly-NMe_2 (**6**), Ac-azaGly-NMe_2 (**7**), and Ac-NMe-azAla-NMe_2 (**8**).

Methyl group substitution at NH positions of azaamino acids incorporated in peptides, as shown in Table 2, would severely perturb the typical β-turn conformation of the azapeptide, and this fact gives rise curiosity about the structural perturbation of azapeptides by methyl group substitution in azaamino acids. Understanding the effects of such alkylation of azaamino acids on the conformation of azapeptides is critical for the design of conformationally bioactive azapeptides. To investigate the influence of structural changes in azapeptides by methyl group substitution in three N-positions of an azaamino acid [i.e., Ac-azaAla-NHMe (**2**), Ac-NMe-azaGly-NHMe (**3**), Ac-NMe-azaAla-NHMe (**4**), Ac-azaGly-NMe_2 (**5**), Ac-azaAla-NMe_2 (**6**), Ac-NMe-azaGly-NMe_2 (**7**), and Ac-NMe-azaAla-NMe_2(**8**)], we

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Table 2. Single-Crystal and Solution Structures of Various Azapeptide Derivatives

azapeptide derivatives	phase	ω_0	ϕ_1	ψ_1	ω_1	structure
(I) Azapeptides Including Primary Amine						
Ac-Aib-**AzaGly**-NH_2	NMR[a]	178	−78	−10	178	βI
Boc-AzaPhe-**AzaGly**-X-AzaIle-OMe[b]	X-ray[c]	−174	65	−168	169	
Boc-**AzaPhe**-AzaGly-X-AzaIle-OMe[b]	X-ray[c]		−109	11	−174	
Boc-AzaPhe-AzaGly-X-**AzaIle**-OMe[b]	X-ray[c]	169	−87	−4	−170	
Piv-Pro-**AzaAla**-NHiPr	X-ray[d]	180	89	18	169	βII
Piv-Pro-**AzaAsn(Me)**-NHiPr	X-ray[e]	178	81	2	−179	βII
Boc-Phe-**AzaLeu**-Ala-OMe	NMR[f]	175	107	−4	175	βII
Boc-Ala-Phe-**AzaLeu**-Ala-OMe	NMR[g]	176	88	1	176	βII (βI)
Boc-Ala-**(NMe)AzaAla**-Ala-NHiPr	X-ray[h]	14	−107	15	177	βVI
(II) Azapeptides Including Secondary Amines						
Boc-**AzaAla**-Pro-NHiPr	X-ray[e]	172	−67	−18	172	βI
Z-**AzaAsn(Me)**-NMe_2	X-ray[e]	−178	−61	−38	179/−11	
Z-**AzaAsn(Me)**-Pro-NHiPr	X-ray[e]	−179	−52	−36	−178	βI

[a] Reference 19. [b] X is an organic spacer. [c] Reference 22. [d] Reference 11. [e] Reference 12. [f] Reference 20. [g] Reference 21. [h] Reference 13.

employed ab initio MO and density function theories to draw the Ramachandran plot and to calculate the minimum energy conformations on the basis of the structure of a model azapeptide, Ac-azaGly-NHMe (**1**).

The pyramidalization of the N2 atom in azapeptide derivatives involves the hybridization of the nitrogen atom in azaamino acid-containing peptides and leads to changes in the N1−N2 and N2−C3 bond lengths. The planar nitrogen indicates sp^2 character, and the pyramidal nitrogen atom implies more sp^3 character. Thus, the N2−C3 bond length increases as the pyramidality of the N2 atom increases.[11−17] Therefore, it is important to know how *N*-methyl groups affect the pyramidality of nitrogens, since that will affect the rotational barriers of N1−N2 (ϕ_1) and N2−C3 (ψ_1). Also, since it was found that the less favorable cis peptide bond occurs in some cases with proteins, the conformational properties of the peptide or peptidomimetic compounds with a cis peptide bond are considered to be equally important as those of the compounds with a trans peptide bond for their possible application in peptide design, and the conformational properties for both orientations of model compounds **1**−**8** were calculated.[23]

2. Computational Methods

The conformational properties of azaamino acid derivatives were investigated using the GAUSSIAN program package run on the Cray T3E supercomputer.[24] The starting structures for ab initio and DFT calculations of azapeptide derivatives **1**−**8** were obtained using two different methods: the potential energy surface and the quenched molecular dynamics.[25]

The potential energy surfaces of compounds **1**−**8** were generated for the two cases with the acetyl group oriented in a trans ($\omega_0 \approx 180°$) or cis ($\omega_0 \approx 0°$) configuration. Relaxed potential energy surfaces were generated on a 30° grid by full optimization of all degrees of freedom except ϕ_1 and ψ_1 at the HF/6-31G*//HF/6-31G* level for **1**−**4** and at the HF/6-31G*//HF/3-21G level for **5**−**8**.[26] The reason molecules **1**−**4** are studied at a different level than molecules **5**−**8** is that for **5**−**8**, due to molecular size, it is appropriate to optimize at first at the HF/3-21G level by rotating two key backbone angles (ϕ, ψ) in increments of 30°, and then perform a single-point energy calculation at the HF/6-31G* level of theory. Numerous studies have shown that the HF/6-31G* level is sufficient to provide an initial picture of the conformational preferences in peptides.[6b,27] Minimum energy conformations were identified by complete geometry optimization of the starting structures selected from the HF/6-31G* or HF/6-31G*//HF/3-21G (ϕ_1, ψ_1) maps. The quenched molecular dynamics were additionally applied to search the starting structures. This calculation involved two steps: high-temperature molecular dynamics at 1000 K for 1000 ps, and full minimization of the structures that appeared every picosecond of the molecular dynamics. For these calculations, the DISCOVER program (Molecular Simulations Inc., 9685 Scranton Rd., San Diego, CA 92121-3752) was used. Additionally, to obtain highly diversified conformations, we repeated the same procedures using three different dielectric constants (ϵ = 1, 45, 80), leading to 4000 AMBER-minimized conformations.[28] These conformations were then classified into conformational families by cluster analysis on the basis of torsion angles. The resulting conformations were fully optimized at the HF/3-21G level, and subsequently selected structures were reoptimized at the HF/6-31G* level. The minima obtained by two different approaches led to the same results.

The HF/6-31G* minimum energy conformations for model compounds **1**−**8** were fully reoptimized at the B3LYP/6-31G* level. All of the stationary points were found to be local minima, as indicated by the absence of imaginary frequencies. The vibrational frequencies calculated at the same levels were also used to compute the enthalpy

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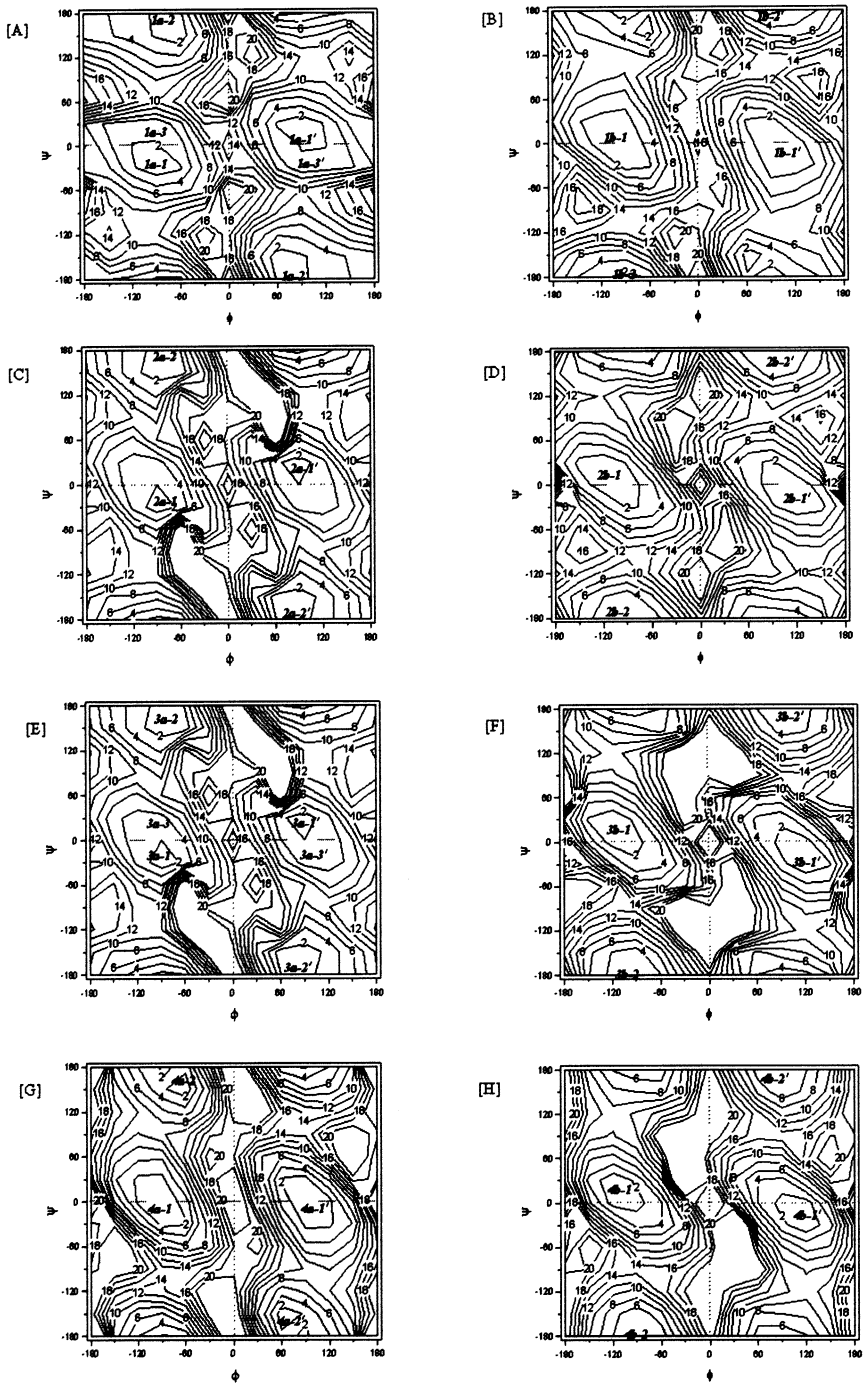

Figure 2. HF/6-31G*//HF/6-31G* potential energy surfaces of azapeptide derivatives (A) **1a**, (B) **1b**, (C) **2a**, (D) **2b**, (E) **3a**, (F) **3b**, (G) **4a**, and (H) **4b**. Here, **a** represents a trans amide orientation ($\omega_0 \approx 180°$) and **b** a cis amide orientation ($\omega_0 \approx 0°$). Geometry optimization of all variables except ϕ_1 and ψ_1 was performed on a grid with 30° spacing. Solid contours are drawn every 2 kcal/mol from the global minimum to 20 kcal/mol. An enlarged minima region for **1a** is available in the Supporting Information.

and the Gibbs energy differences for the cis/trans isomerization at 298 K.[29]

3. Results and Discussion

3.1. Potential Energy Surfaces. The potential energy surfaces for trans and cis orientations of an acetyl amide bond were generated by rotating two key dihedral angles, ϕ_1 and ψ_1, in increments of 30° at the HF/6-31G* level for the compound Ac-azaGly-NHMe (**1**). During the rotation, the acetyl amide bond was fixed with a trans (**1a**−**8a**; $\omega_0 \approx 180°$) or cis (**1b**−**8b**; $\omega_0 \approx 0°$) orientation. On the basis of the HF/6-31G* potential energy surfaces, the starting structures were selected and then fully optimized at the HF and B3LYP levels with the 6-31G* basis set. These potential energy surfaces were generated further for Ac-azaAla-NHMe (**2**), Ac-NMe-azaGly-NHMe (**3**), and Ac-NMe-azaAla-NHMe (**4**) to figure out how the methyl groups affect the conformational preference of those azapeptides in comparison to that of the simplest representative of an azapeptide, **1**. As shown in Figure 2, the potential energy

(29) Kang, Y. K. *J. Mol. Struct. (THEOCHEM)* **2001**, *546*, 183 and references therein.

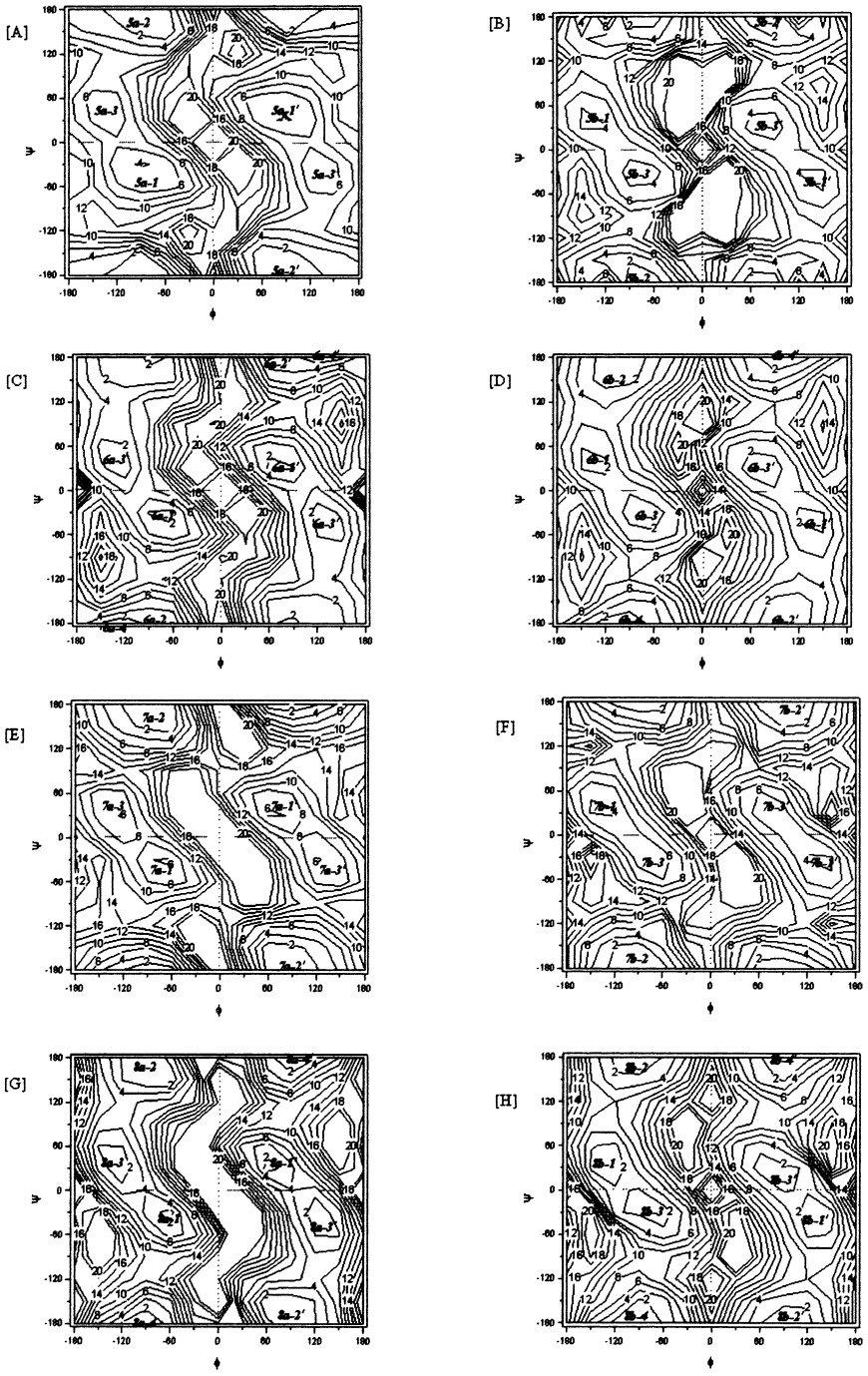

Figure 3. HF/6-31G*//HF/3-21G potential energy surfaces of azapeptide peptide derivatives (A) **5a**, (B) **5b**, (C) **6a**, (D) **6b**, (E) **7a**, (F) **7b**, (G) **8a**, and (H) **8b**. Here, **a** represents a trans amide orientation ($\omega_0 \approx 180°$) and **b** a cis amide orientation ($\omega_0 \approx 0°$). Geometry optimization of all variables except ϕ_1 and ψ_1 was performed on a grid with 30° spacing. Solid contours are drawn every 2 kcal/mol from the global minimum to 20 kcal/mol. An enlarged minima region for **5a** is available in the Supporting Information.

surfaces for azapeptide derivatives **1**−**4** show similar features, giving a C_2 symmetry axis ($\phi_1 \approx 0°$, $\psi_1 \approx 0°$); the replacement of hydrogen by a methyl at the N1 and/or N2 position would not perturb much the structure of Ac-azaGly-NHMe (**1**) published earlier, irrespective of the acetyl amide orientation.[17,18b] However, the curvature patterns of *N*-methyl-substituted azapeptide derivatives are steeper than that of compound **1**, which demonstrates that methyl groups at the N1 or N2 position would restrict the allowed ϕ_1 and ψ_1 regions of the azaamino acid. The low-energy conformations of **1**−**4** are located in the right-handed regions (δ_R; $\phi_1 \approx -90° \pm 30°$, $\psi_1 \approx 0° \pm 30°$) and polyproline II regions (β_P; $\phi_1 \approx -80° \pm 30°$, $\psi_1 \approx \pm 170° \pm 30°$) in the Ramachandran plots. These are the characteristic potential energy surfaces for azaamino acids, and their local minimum energy conformations are not located in the β-sheet regions (β_S; $\phi_1 \approx -180° \pm 30°$, $\psi_1 \approx 180° \pm 30°$), in contrast to those of natural amino acids.[27,30]

The potential energy surfaces of compounds Ac-azaGly-NMe_2 (**5**), Ac-azaAla-NMe_2 (**6**), Ac-NMe-azaGly-NMe_2 (**7**), and Ac-NMe-azaAla-NMe_2 (**8**) coupled to a secondary amine were also

(30) Notation: Karplus, P. A. *Protein Sci.* **1996**, *5*, 1406.

generated. Due to the molecular size, the HF/3-21G optimizations for **5**−**8** were performed by rotating two key backbone (ϕ, ψ) angles in increments of 30°. On the basis of these HF/3-21G-optimized structures, a single-point energy calculation was performed at the HF/6-31G* level of theory. As shown in Figure 3, the potential energy surfaces for compounds **5**−**8** also have a symmetric counterpart, but the potential energy surfaces of compounds **5**−**8** show significantly different features as compared to those of **1**−**4**. The local minimum energy conformations for compound **5** were found to be located at the polyproline II (β_P; $\phi_1 \approx -80°$, $\psi_1\ \theta \approx 170°$), the bend ($\zeta$; $\phi_1 \approx -120°$, $\psi_1 \approx 60°$), and the α-helical (α_R; $\phi_1 \approx -60°$, $\psi_1 \approx -30°$) regions, regardless of the orientation of the acetyl amide group. The similarity of the potential energy surfaces for compounds **6**−**8** with that of compound **5** indicates that the *N*-methyl groups at the N1 and N2 positions do not perturb the local minimum energy conformations for **5** but restrict the energetically allowed region of the conformers. It is noticed that the β-strand regions (β_S; $\phi_1 \approx 180°$, $\psi_1 \approx 180°$) were destabilized by *N*-methylation.

3.2. The Rotational Energy Barriers. The conformational preferences of azapeptide derivatives have been known to be related to the repulsion between nitrogen lone pairs and the conjugated amide bond in the urea-type structure.[18,31] This fact can be rationalized on the basis of the rotational barriers of the N1−N2 (ϕ_1) and N2−C3 (ψ_1) bonds, estimated on the basis of the potential energy surfaces in Figures 2 and 3; these rotational barriers are shown in Table 3. These rotational barriers could give insight into the preferred conformation or rigidity of azapeptides. The barriers of the N1−N2 bond for rotation through path I (rotation of ϕ_1 from the minimum through −180°) or path II (rotation of ϕ_1 from the minimum through 0°) were estimated at fixed ψ_1 (0° or 180°). The rotational barriers of the N1−N2 bond range from 2.9 to 35.0 kcal, depending on the orientation of the acetyl amide group and the number of methyl groups bound to azapeptide derivatives. Methyl groups at the N3 position would not affect the rotational barriers of the N1−N2 bond, but methyl groups bound at N1 and N2 positions increase its rotational barriers due to steric hindrance, as presented in Table 3. The barriers of the N2−C3 bond for rotation through path III (rotation of ψ_1 from the minimum through −180°) or path IV (rotation of ψ_1 from the minimum to 180°) were estimated at fixed ϕ_1 (−90°). The rotational barriers of the N2−C3 bond seem not to depend on the orientation of the acetylamide group, and the average rotational barrier for the N2−C3 bond of azapeptides **1**−**4** is about 11.0 kcal/mol, which is relatively higher than that (8.0 kcal/mol) of azapeptides **5**−**8**, which means that the additional methyl group at N3 lowers the rotational barriers for the N2−C3 bond, as shown in Table 3. These rotational energy barriers, as functions of conformers and configurations, allow us to visualize the influence of *N*-methyl substitutions on the conformations of azapeptides.

3.3. Minimum Energy Conformations. As shown in the Ramachandran plots in Figures 2 and 3, the minimum energy conformations for **1**−**8** have their own mirror image conformations, and only one symmetric part of the mirror image conformations is considered. The resulting B3LYP/6-31G*-optimized structures, including the geometries and intramolecular hydrogen-bonding parameters for **1**−**8**, are displayed in Figures 4−11. The B3LYP/6-31G* backbone (ϕ_1, ψ_1) angles and the relative energies, enthalpies, and Gibbs energies of minima for **1**−**8** are given in Tables 4 and 5.

Table 3. Rotational Barriers (kcal/mol) for the N1−N2 Bond (ϕ) and N2−C3 Bond (ψ) of Azapeptide Derivatives **1**−**4** at the HF/6-31G* Level and **5**−**8** at the HF/6-31G*//HF/3-21G Level

N1−N2 Bond (ϕ)

	path **I** (min → $\phi = -180°$)		path **II** (min → $\phi = 0°$)	
	fixed angle $\psi = 0°$	fixed angle $\psi = 180°$	fixed angle $\psi = 0°$	fixed angle $\psi = 180°$
1a	6.4	2.9	14.1	21.5
1b	11.2	6.5	16.8	24.4
2a	12.7	8.4	17.6	24.7
2b	21.5	14.8	22.9	12.9
3a	12.7	8.4	17.6	24.7
3b	17.5	15.9	25.3	15.4
4a	27.1	36.1	26.1	25.6
4b	23.9	18.8	28.8	17.3
5a	4.6	3.2	23.6	22.3
5b	9.3	13.6	21.7	13.2
6a	23.4	5.7	25.8	24.8
6b	12.1	9.1	20.4	14.6
7a	9.4	9.8	27.3	25.1
7b	14.2	12.8	23.5	16.0
8a	24.9	19.6	35.0	15.2
8b	21.2	17.5	24.6	16.5

N2−C3 Bond (ψ)

	path **III** (min → $\psi = -180°$)	path **IV** (min → $\psi = 180°$)
	fixed angle $\phi = -90°$	fixed angle $\phi = -90°$
1a	11.0	9.2
1b	12.6	6.9
2a	11.0	9.1
2b	10.2	12.1
3a	11.0	9.1
3b	13.8	11.6
4a	14.2	9.5
4b	14.1	11.2
5a	9.7	6.5
5b	5.9	10.0
6a	9.0	5.0
6b	7.0	6.5
7a	9.4	8.4
7b	10.8	8.9
8a	8.7	5.8
8b	8.1	7.3

Ac-azaGly-NHMe (1). Five minima were found at the B3LYP/6-31G* level, as presented in Table 4 and Figure 4. Three minima for compound **1a** ($\omega_0 \approx 180°$) were characterized. The backbone angles for all minima were found in the secondary structure of native proteins or peptides, which indicates that the incorporation of azaamino acids in peptides could stabilize the specific conformations. Conformer **1a-1** (δ_R; $\phi_1 = -94°$, $\psi_1 = -24°$) adopts a βI(I′)- or βII(II′)-turn in the $i + 2$ position, and conformer **1a-2** (β_P; $\phi_1 = -73°$, $\psi_1 = 165°$) appears to be the polyglycine II or polyproline II helix structure, as displayed in Table 1. Conformer **1a-1** and the mirror image of **1a-2** were found in solution as well as in the solid state (see Table 2). Conformer **1a-3** (δ_R/γ'; $\phi_1 = -82°$, $\psi_1 = 37°$) has a backbone angle similar to that of **1a-1**, but a distinct difference is seen in the pattern of the intramolecular hydrogen bond. The hydrogen bond of conformer **1a-1** forms a five-membered ring involving the nitrogen lone pair, N1(lp), and the H−N3 bond, whereas

(31) (a) Reynolds, C. H.; Hormann, R. E. *J. Am. Chem. Soc.* **1996**, *118*, 9395 and references therein. (b) Alemán, C.; Puiggalí, J. *J. Org. Chem.* **1999**, *64*, 351.

Table 4. Dihedral Angles and Relative Energies (ΔE at 0 K, in kcal/mol), Enthalpy (ΔH at 298 K, in kcal/mol), and Gibbs Energies (ΔG at 298 K, in kcal/mol) for the Minima of Azapeptide Derivatives **1**−**4** at the B3LYP/6-31G* Level

conformer	ω_0	ϕ_1	ψ_1	ω_1	ΔE	ΔE^a	ΔH	ΔG	$\sum$N2[b]	structure[c]
1a-1	−168	−94	−24	178	0.22	0.51	0.51	0.37	341.0	δ_R
1a-2	166	−73	165	171	1.18	1.42	1.23	0.71	337.4	β_P
1a-3	179	−82	37	173	1.92	2.17	2.35	2.78	348.6	δ_R/γ'
1b-1	15	−125	20	176	**0.00**	0.00	0.00	0.00	349.2	δ_R
1b-2	−19	−95	−169	−172	2.30	2.77	2.42	2.31	345.0	ϵ'
2a-1	−167	−92	−13	173	1.36	1.44	1.37	0.91	355.9	δ_R
2a-2	163	−71	163	−178	2.57	2.31	3.05	3.50	354.5	β_P
2b-1	19	−119	15	−179	**0.00**	0.00	0.00	0.00	353.6	δ_R
2b-2	−19	−82	−167	−171	4.32	4.58	4.27	4.20	352.8	ϵ'
3a-1	−171	−94	−22	176	3.10	3.11	3.19	3.06	341.3	δ_R
3a-2	175	−84	166	172	2.98	3.03	3.20	2.64	334.1	β_P
3a-3	−175	−87	40	172	4.27	4.45	4.99	5.77	345.2	δ_R/γ'
3b-1	12	−122	23	174	**0.00**	0.00	0.00	0.00	346.9	δ_R
3b-2	−8	−93	−165	178	2.60	2.86	2.59	2.65	347.1	ϵ'
4a-1	−171	−92	−9	173	3.42	3.28	3.66	3.91	357.0	δ_R
4a-2	172	−70	160	−178	4.13	4.02	4.77	6.06	352.6	β_P
4b-1	12	−114	15	174	**0.00**	0.00	0.00	0.00	354.1	δ_R
4b-2	−10	−84	−165	178	3.78	3.93	3.91	4.55	356.7	ϵ'

[a] B3LYP/6-311++G**//B3LYP/6-31G*. [b] The sum of the bond angles around nitrogen. [c] The notations are according to ref 30.

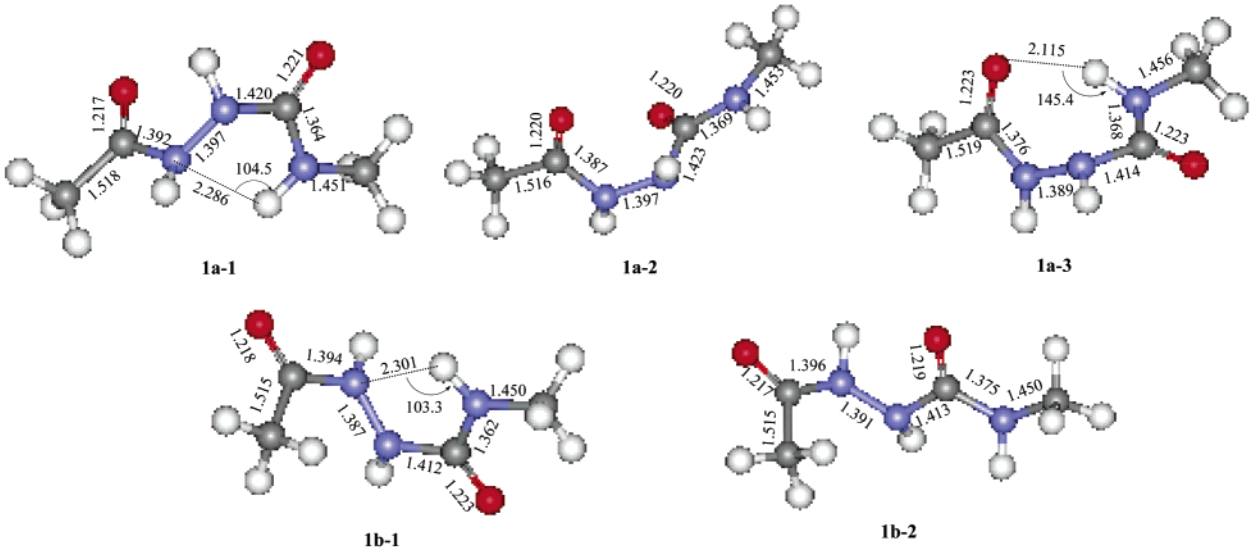

Figure 4. B3LYP/6-31G* minima of Ac-azaGly-NHMe (**1**). The geometries with intramolecular hydrogen-bonding parameters are included.

the hydrogen bond of conformer **1a-3** forms a seven-membered ring between the C2═O1 and N3—H bonds (r_{O1-H} = 2.115 Å, ∠N3—H- - -O1 = 145.4°). A γ-turn-like conformation in **1a-3** by a hydrogen bond would not stabilize its conformation, since ψ_1 deviates from the ideal γ-turn structure ($\phi_1 \approx -80°$, $\psi_1 \approx 70°$) of natural peptides defined by the characteristic N2—C3 bond angle. Although this H-bond angle (104.5°) of **1a-1** is relatively small, this type of the intramolecular H-bond was recognized to stabilize the conformation.[20,21] The minimum energy conformations for **1a** are generally consistent with previous results.[17,20] Two minima for **1b** ($\omega_0 \approx 0°$) were generated. The backbone angle of conformer **1b-1** (δ_R; $\phi_1 = -125°$, $\psi_1 = 20°$) is found at the $i+2$ position of the βVIa-turn, and conformer **1b-2** (ϵ'; $\phi_1 = -95°$, $\psi_1 = -169°$) adopts an extended conformation. The hydrogen bond of conformer **1b-1** forms a five-membered ring, which is similar to the case with conformer **1a-1**. The lowest energy conformation for **1** is determined to be conformer **1b-1** at the B3LYP/6-31G* level or by a single-point energy calculation at the B3LYP/6-31++G**//B3LYP/6-31G* level, as displayed in Table 4. The relative enthalpy and Gibbs energy also indicate that **1b-1** is the global minimum. This fact is unique, from the point of view that a trans peptide bond is more favorable than a cis peptide bond in natural peptides.[32] Since the relative energy of **1b-1** is a little lower than that of **1a-1**, two isomers could coexist in solution with a higher proportion of **1b-1**. Thus, model **1** could be utilized as the βVIa-turn scaffold, as in the Xaa-azaPro sequence, where Xaa is an amino acid.[13,14,18b]

Ac-azaAla-NHMe (2). As shown in Table 4 and Figure 5, four minima are generated with detailed geometries and intramolecular hydrogen-bonding parameters. The backbone angles of minima for **2** are very close to those for **1** except that the γ-turn-like conformer in **2a** was not a local minimum. The most stable conformation for **2** is shown by conformer **2b-1**, which adopts a backbone angle similar to that for **1**. These results indicate that *N*-methyl substitution on the N2 atom does not alter the conformational properties and may stabilize the cis peptide bond. Conformers **2b-1** (δ_R; $\phi_1 = -119°$, $\psi_1 = 15°$) and **2a-1** (δ_R; $\phi_1 = -92°$, $\psi_1 = -13°$) would be stabilized by the intramolecular H-bond interaction of N(lp)- - -H—N. The

(32) The energy difference between native peptides with cis and trans amide bonds is about 2.87−3.11 kcal/mol.

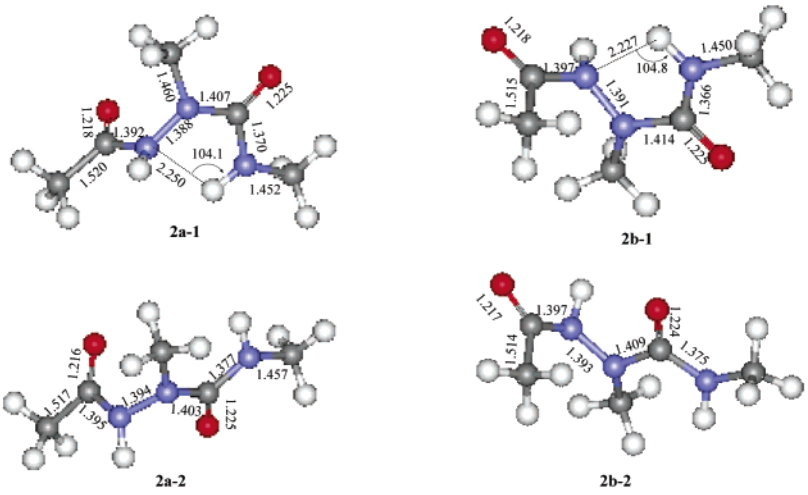

Figure 5. B3LYP/6-31G* minima of Ac-azaAla-NHMe (**2**). The geometries with intramolecular hydrogen-bonding parameters are included.

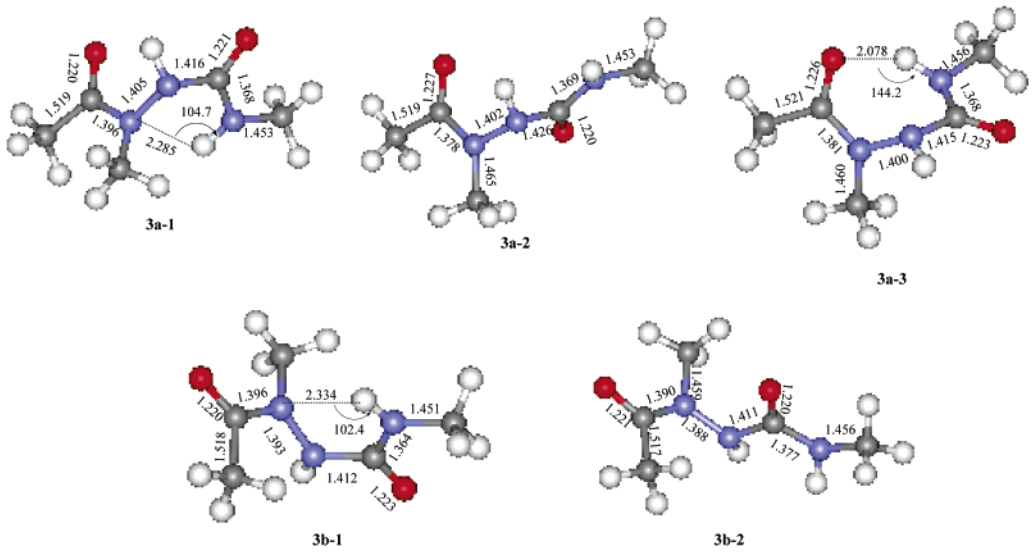

Figure 6. B3LYP/6-31G* minima of Ac-NMe-azaGly-NHMe (**3**). The geometries with intramolecular hydrogen-bonding parameters are included.

relative energy difference between **2b-1** and **2a-1** is in the range of 1.36−1.44 kcal/mol, depending on the calculation method used, and the relative Gibbs energy difference between **2a-1** and **2b-1** is small, 0.91 kcal/mol at the B3LYP/6-31G* level. However, the X-ray and NMR structures shown in Table 2 indicate that azaalanine-containing peptides adopt the backbone angles of **2a-1** and its enantiomer rather than those of **2b-1**. This indicates that the calculated minimum energy conformers may not be the real minima in solution or in the solid state, although the substituents of azaAla-containing peptides in Table 2 are different from those of **2**.

Ac-NMe-azaGly-NHMe (3). Five minima were generated, as shown in Figure 6 and Table 4. These minima for **3** are very close to those for **1**. As discussed for **1**, conformer **3b-1** (δ_R; $\phi_1 = -122°$, $\psi_1 = 23°$), forming a five-membered ring with a hydrogen bond ($r_{N1-H} = 2.334$ Å, ∠N3−H- - -N1 = 102.4°), is the lowest energy conformation. Conformer **3a-2** (β_P; $\phi_1 = -84°$, $\psi_1 = 166°$), appearing to have the polyproline II or polyglycine II structure, is second only to **3b-1** in relative energy. The difference in energy between conformers **3b-1** and **3a-2** is 3.03 or 3.20 kcal/mol at the two levels of theory. The relative enthalpy and Gibbs energy for the two conformers are found to be 3.20 and 2.64 kcal/mol, respectively. The results indicate that the cis peptide bond is stabilized by *N*-methyl substitution on the N1 atom. Although it was not certain that the calculated minimum conformers can be conserved in solution or in the solid state, there is some evidence that the theoretical minimum conformers for azapeptides containing a N(Me)-azaAla residue are consistent with those in solution or the solid state. This will be shown for the minima for **4** (see below).

Ac-NMe-AzaAla-NHMe (4). The backbone angles of minima for **4** are very close to those for **2**, which indicates that the conformational properties for **2** and **4** are essentially the same (see Figures 5 and 7). Conformer **4b-1** (δ_R; $\phi_1 = -114°$, $\psi_1 = 15°$) is the most stable in terms of the relative energy, enthalpy, and free energy, as shown in Table 4. Therefore, it is found that all of the minimum energy conformers for **1**−**4** adopt the cis amide bond. However, this may not always be true. Didierjean et al. reported that the crystal structure of Boc-Ala-NMe-azaAla-Ala-NHiPr with a N(Me)-azaAla residue adopts the βVIa-turn ($\omega_{i+1} = 14°$, $\phi_{i+2} = -107°$, $\psi_{i+2} = 15°$) like **4b-1**, and this model compound in solution was found to coexist in a ratio of 80(cis):20(trans) in $CDCl_3$ and 25(cis):75(trans) in

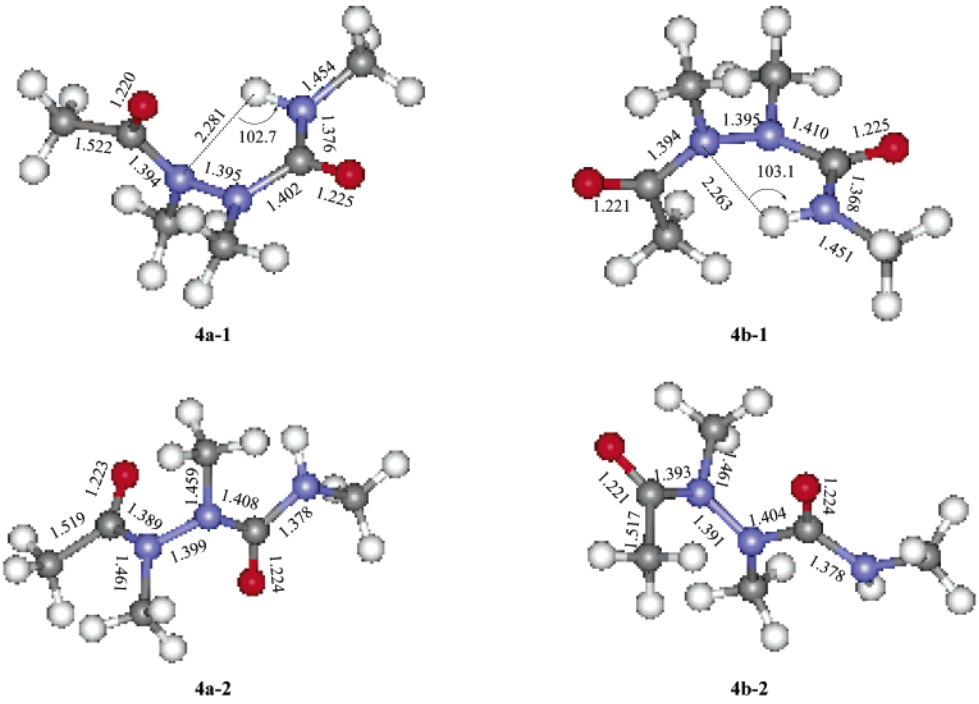

Figure 7. B3LYP/6-31G* minima of Ac-NMe-azaAla-NHMe (**4**). The geometries with intramolecular hydrogen-bonding parameters are included.

Table 5. Dihedral Angles and Relative Energies (ΔE at 0 K, in kcal/mol), Enthalpy (ΔH at 298 K, in kcal/mol), and Gibbs Energies (ΔG at 298 K, in kcal/mol) for the Minima of Azapeptide Derivatives **5**−**8** at the B3LYP/6-31G* Level

conformer	ω_0	ϕ_1	ψ_1	ω_1	ΔE	ΔE^a	ΔH	ΔG	ΣN2[b]	structure[c]
5a-1	−170	−83	−46	174	3.36	3.69	3.38	3.57	335.7	α_R
5a-2	165	−75	168	−175	0.00	0.00	0.00	0.00	336.9	β_P
5a-3	−172	−131	19	−175	3.83	3.72	3.88	4.48	341.7	δ_R
5b-1	10	−144	44	−174	3.97	3.75	3.84	4.17	338.9	ζ
5b-2	−20	−103	−174	172	1.39	1.69	1.22	1.49	342.1	ϵ'
5b-3	10	−73	−45	174	3.67	3.54	3.62	3.92	343.5	α_R
6a-1	−169	−67	−41	172	1.34	1.95	1.03	1.02	346.5	α_R
6a-2	161	−71	−175	−173	2.33	2.80	2.31	2.52	344.0	ϵ'
6a-3	−169	−135	46	−173	0.11	0.50	0.00	0.19	347.6	ζ
6a-4	170	−134	−172	173	0.00	0.00	0.03	0.00	351.4	β_S
6b-1	12	−141	44	−172	1.61	1.78	1.42	1.69	343.5	ζ
6b-2	−19	−106	168	173	1.24	1.75	0.86	0.95	337.7	ϵ''
6b-3	14	−72	−41	172	1.52	1.67	1.46	1.51	351.6	α_R
6b-4	−18	−67	−166	−169	3.49	4.06	3.36	3.94	358.3	ϵ'
7a-1	−173	−66	−43	174	5.16	5.24	5.36	5.95	336.6	α_R
7a-2	174	−83	169	−175	0.00	0.00	0.36	0.36	333.8	β_P
7a-3	−172	−132	48	−175	4.82	4.79	5.19	5.94	341.4	ζ
7b-1	8	−142	43	−175	2.18	2.08	2.37	2.71	345.6	ζ
7b-2	−8	−91	−170	175	0.15	0.41	0.00	0.00	347.7	ϵ'
7b-3	14	−68	−40	175	3.45	3.14	3.48	3.65	338.7	α_R
8a-1	−173	−63	−40	171	2.54	2.92	2.35	2.78	345.6	α_R
8a-2	175	−83	156	165	1.35	2.61	1.61	1.96	359.7	β_P
8a-3	−173	−131	38	−173	0.97	1.24	1.14	1.48	352.0	ζ
8a-4	169	−74	−175	−172	2.29	1.62	2.52	3.23	351.8	ϵ'
8b-1	5	−134	40	−173	0.00	0.00	0.00	0.00	352.4	ζ
8b-2	−4	−95	173	172	1.25	1.62	1.16	1.51	359.8	ϵ''
8b-3	16	−69	−36	173	0.70	0.61	0.52	0.32	345.7	α_R
8b-4	−11	−76	−164	−167	0.79	0.95	0.71	0.63	353.5	ϵ'

[a] B3LYP/6-311++G**//B3LYP/6-31G*. [b] The sum of the bond angles around nitrogen. [c] The notations are according to ref 30.

DMSO-d_6.[13,14] These facts suggest that the lowest energy conformers for azapeptide derivatives can be changed as a function of solvent.

Ac-azaGly-NMe$_2$ (5). Six minimum energy conformations were generated at the B3LYP/6-31G* level, as presented in Table 5 and Figure 8. The minima for **5** coupled to a secondary amine are significantly different from those for **1** coupled to a primary amine at the C-terminus. This demonstrates that the *N*-methyl group substituent on the N3 atom could significantly change the conformational behaviors of azapeptides. The lowest energy conformation in **5** is the trans conformer **5a-2** (β_P; $\phi_1 = -75°$, $\psi_1 = 168°$), appearing to be the polyproline II or polyglycine II structure, whereas the conformers with a cis peptide bond in **1**−**4** were the global minima. Interestingly, conformer **5a-1** (α_R; $\phi_1 = -83°$, $\psi_1 = -46°$), appearing to have the βI(III) structure in the $i + 1$ position, is found as a

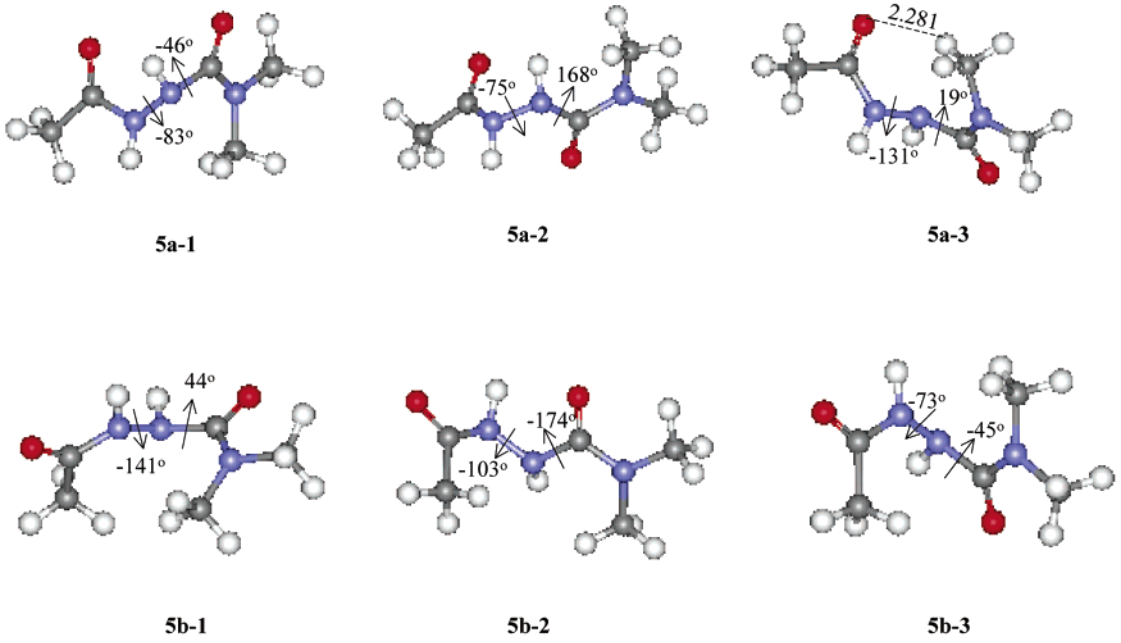

Figure 8. B3LYP/6-31G* minima of Ac-azaGly-NMe_2 (**5**). The backbone angles are included.

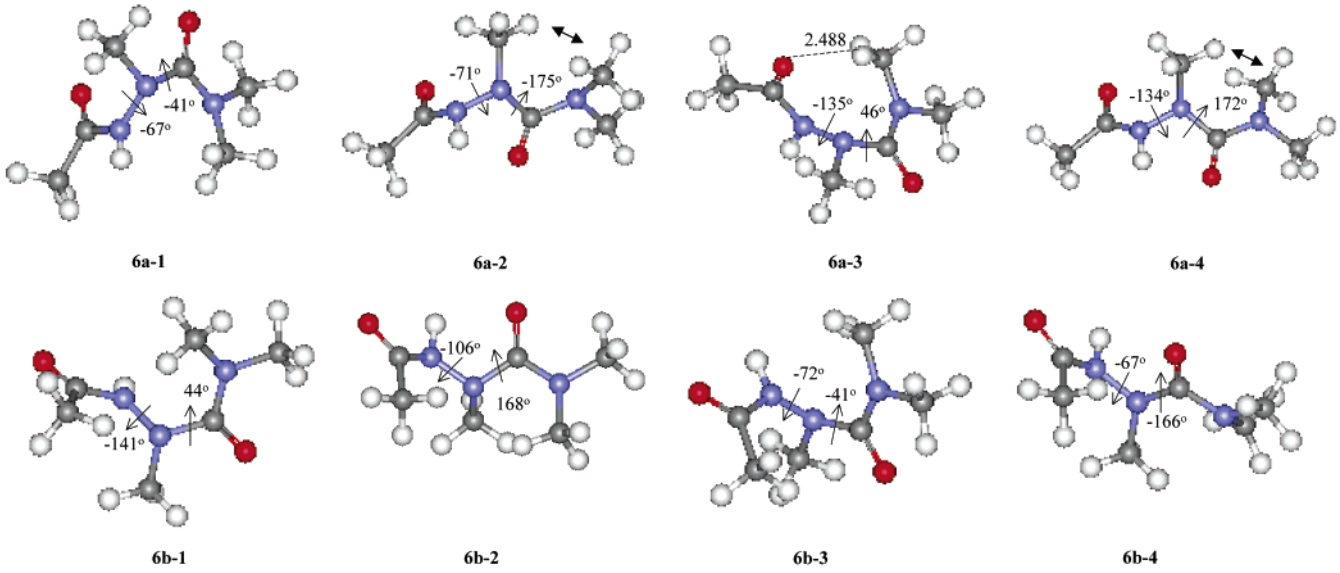

Figure 9. B3LYP/6-31G* minima of Ac-azaAla-NMe_2 (**6**). The backbone angles are included.

local minimum which is 3.36 kcal/mol less stable than **5a-2**. The ψ_1 angle of **5a-1**, with an additional *N*-methyl group substituent on the N3 atom, is distorted by about 20° with respect to that of **1a-1**. Conformer **5a-3** (δ_R; $\phi_1 = -131°$, $\psi_1 = 19°$) forms a seven-membered ring with a weak hydrogen bond between C2═O1 and the hydrogen of the N3-methyl group ($r_{O1\cdots H} = 2.452$ Å, ∠O1- -H−C = 150.0°).

As shown in Figure 8, the minima of **5b** with a cis peptide bond show conformational behavior similar to to that of **5a** with a trans peptide bond. Note that conformer **5b-3** (α_R; $\phi_1 = -73°$, $\psi_1 = -45°$), with a helix structure, appears to be a local minimum which is 3.67 kcal/mol less stable than the global minimum.

Ac-AzaAla-NMe_2 (6). Eight minimum energy conformations were generated, as shown in Table 5 and Figure 9. The backbone (ϕ, ψ) angles of minima for compound **6** were greatly altered with respect to those for compound **2** and are similar to those for **5**. Conformer **6a-1** has a backbone angle of $\phi_1 = -67°$ and $\psi_1 = -41°$ corresponding to the α-helix or the $i + 1$ position of and ideal β-I(III)-turn structure, consistent with a single-crystal structure (Table 2). The conformer with the most stable conformation is **6a-4** (β_S; $\phi_1 = -134°$, $\psi_1 = 172°$), located at the boundary of the β-sheet (β_S) and extended (ϵ') structures, which is not observed in **2** and **5**. Although conformer **6a-3** (ζ; $\phi_1 = -134°$, $\psi_1 = 46°$) is predicted to be the most stable in terms of enthalpy energy, Gibbs energy of conformer **6a-4** is the global minimum. The energy difference between conformers **6a-3** and **6a-4** is about 0.0−0.5 kcal/mol, depending on the calculation method used. As shown in Figure 9, the difference in the backbone dihedral angles between conformers **6a-2** (ϵ'; $\phi_1 = -71°$, $\psi_1 = -175°$) and **6a-4** is due to the difference in the steric repulsion, according to the orientation of the methyl groups on the N2 and N3 atoms. Conformer **6a-3** (ζ; $\phi_1 = -135°$, $\psi_1 = 46°$), like **5a-3**, forms a seven-membered ring

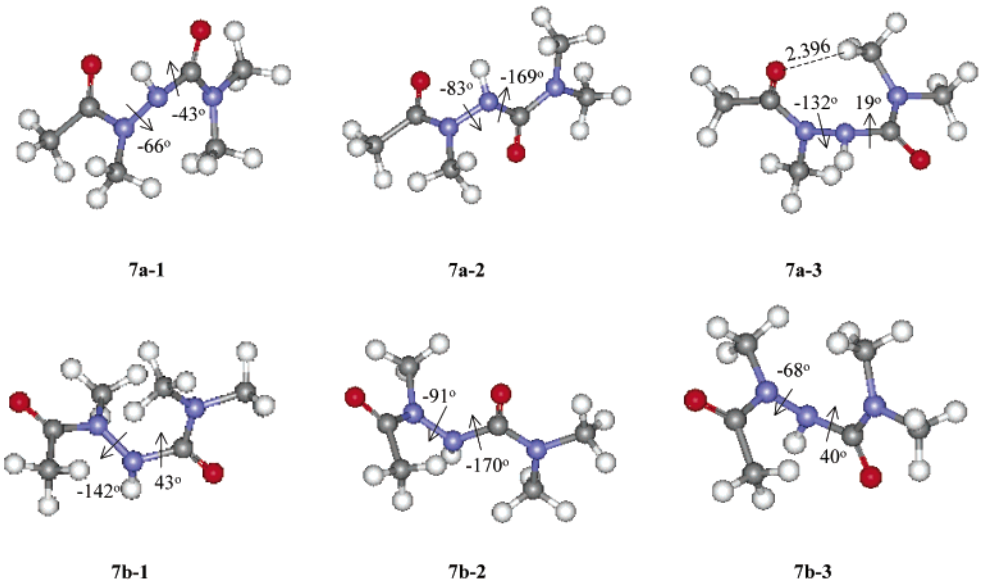

Figure 10. B3LYP/6-31G* minima of Ac-NMe-azaGly-NMe$_2$ (**7**). The backbone angles are included.

with a hydrogen bond between C2═O1 and the C-terminal methyl group (Figure 9). Conformer **6b-3** (α_R; $\phi_1 = -72°$, $\psi_1 = -41°$), with helical structure like **5b-3**, is found as a local minimum. The energy difference between conformer **6b-3** and the global minimum **6a-4** is 1.52 kcal/mol at the B3LYP/6-31G* level. A single-point energy calculation shows that the helical conformer **6a-1** is 0.18 kcal/mol more stable than the helical conformer **6b-3** at the B3LYP/6-311++G**//B3LYP/6-31G* level.

Ac-NMe-AzaGly-NMe$_2$ (7). Six minimum energy conformations were generated at the B3LYP/6-31G* level, as presented in Table 5 and Figure 10. The backbone angles of minima for **7** are similar to those for **5**. Conformer **7a-2** (β_P; $\phi_1 = -83°$, $\psi_1 = 169°$), adopting the polyproline II structure, is the lowest energy conformer, although conformer **7b-2** (ϵ'; $\phi_1 = -91°$, $\psi_1 = -170°$) is the most stable in terms of the enthalpy and Gibbs energy. However, the two conformers could coexist in solution due to the small energy difference. Conformer **7a-3** (ζ; $\phi_1 = -132°$, $\psi_1 = 48°$) forms a seven-membered ring with a hydrogen bond like conformers **5a-3** (δ_R) and **6a-3** (ζ). These facts indicate that the methyl groups bound to N1 and N2 atoms would not perturb their own backbone skeleton by much.

Moehle and Hofmann reported earlier three minima for Ac-NMe-Gly-NMe$_2$ with a trans or a cis peptide bond at the MP2/6-31G* level.[33] The backbone dihedral angles for Ac-NMe-Gly-NMe$_2$ with a trans peptide bond were (α_R; $\phi_1 = -55°$, $\psi_1 = -47°$), (β_P; $\phi_1 = -74°$, $\psi_1 = 176°$), and (ζ; $\phi_1 = -128°$, $\psi_1 = 77°$), and those for Ac-NMe-Gly-NMe$_2$ with a cis peptide bond were (ζ; $\phi_1 = -158°$, $\psi_1 = 63°$), (ϵ'; $\phi_1 = -72°$, $\psi_1 = -172°$), and (α_R; $\phi_1 = -62°$, $\psi_1 = -51°$). Although the backbone angles of the minima for **7** are similar to those of Ac-NMe-Gly-NMe$_2$, the relative energies of the minima for **7** are significantly different from those of the minima for Ac−NMe-Gly-NMe$_2$. For example, the global minimum for **7** and Ac-NMe-Gly-NMe$_2$ adopts β_p and ζ conformers, respectively. In particular, the local minimum energy of helical structures for **7** is 5.2 kcal/mol less stable than its global minimum energy, and the local minimum energy of the helical structure for Ac-NMe-Gly-NMe$_2$ is 17−24 kcal/mol less stable than its global minimum. These facts indicate that the conformational properties for **7** are different from those of Ac-NMe-Gly-NMe$_2$.

Ac-NMe-AzaAla-NMe$_2$ (8). Eight minima were generated at the B3LYP/6-31G* level, as presented in Table 5 and Figure 11. The backbone dihedral angles of the minima for **8** are similar to those for **6**, which confirmed that *N*-methyl substitution on the N1 atom would not perturb the conformations, as suggested earlier, but the relative energy order of the minima for **8** was not consistent with those of **6**. The lowest energy conformation is conformer **8b-1** (ζ; $\phi_1 = -134°$, $\psi_1 = 40°$) in terms of the enthalpy and Gibbs energy as well as the relative energy. The helical conformer **8b-3** (α_R; $\phi_1 = -69°$, $\psi_1 = -36°$), in terms of relative energies, is second only to the lowest energy conformer, **8b-1**. The energy difference between **8b-1** and **8b-3** is about 0.61 kcal/mol at the B3LYP/6-311++G**//B3LYP/6-31G* level of theory. Remarkably, the helical conformer **8a-1** (α_R; $\phi_1 = -63°$, $\psi_1 = -40°$) is about 2 kcal/mol less stable than conformer **8b-3**. This shows that the helical structure with a cis peptide bond in **8** is stabilized by *N*-methyl substitution on the N1 atom, as compared to the helical structures in **6**. As discussed for compound **6**, the structures of the trans conformers **8a-2** (β_P; $\phi_1 = -83°$, $\psi_1 = 156°$) and **8a-4** (ϵ'; $\phi_1 = -74°$, $\psi_1 = -175°$) and corresponding cis conformers **8b-2** (ϵ''; $\phi_1 = -95°$, $\psi_1 = 173°$) and **8b-4** (ϵ'; $\phi_1 = -76°$, $\psi_1 = -164°$) seem to be related in terms of the minimization of the steric repulsion between the methyl groups of the N2 and N3 atoms. The relative energy difference between the different minima for **8** is rather small, in the range of 0.7−2.5 kcal/mol at the B3LYP/6-31G* level. This implies that the structures for **8** could coexist as diverse conformers in solution.

3.4. Nitrogen Pyramidalization and Bond Length. The characteristic structural difference, in addition to the backbone dihedral angles, in *N*-methyl azapeptide derivatives is the pyramidalization of the N2 position (see Tables 4 and 5). The average ∑N2 values, indicating the sum of bond angles around the N2 atom, range from 339.8° to 344.2° in compounds **1**, **3**, **5**, and **7**, including the azaGly residue, whereas those of compounds **2**, **4**, **6**, and **8**, including the azaAla residue, range

(33) Moehle, K.; Hofmann, H.-J. *Biopolymers* **1996**, *38*, 781.

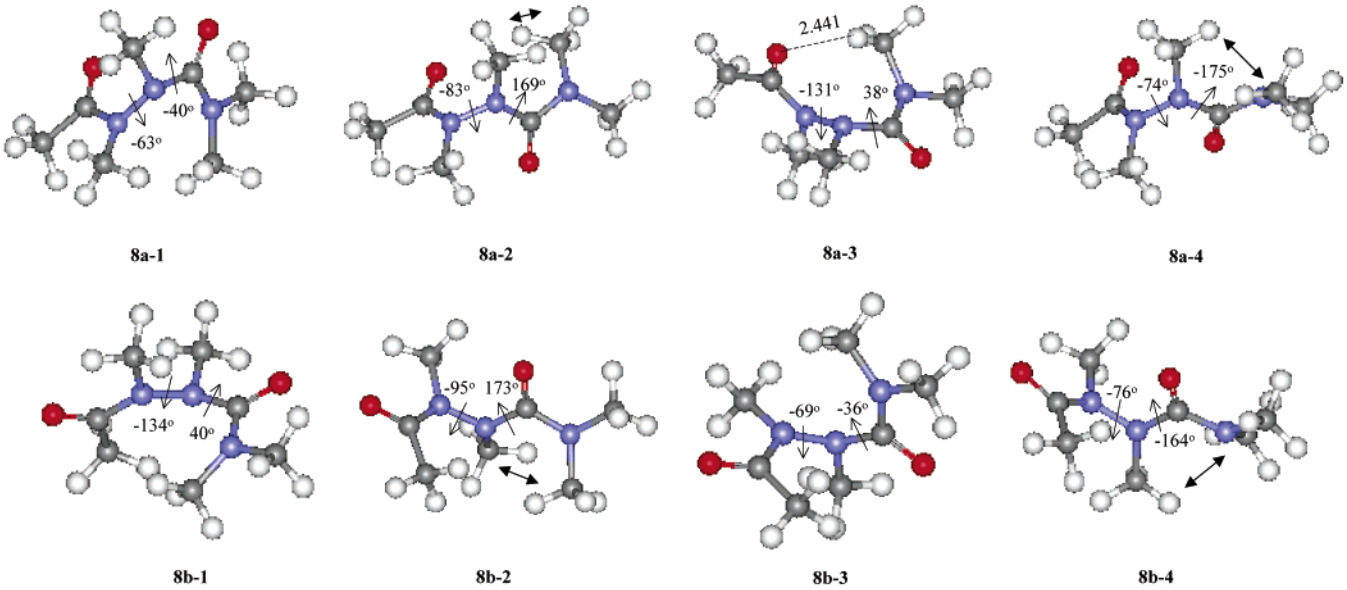

Figure 11. B3LYP/6-31G* minima of Ac-NMe-azaAla-NMe$_2$ (**8**). The backbone angles are included.

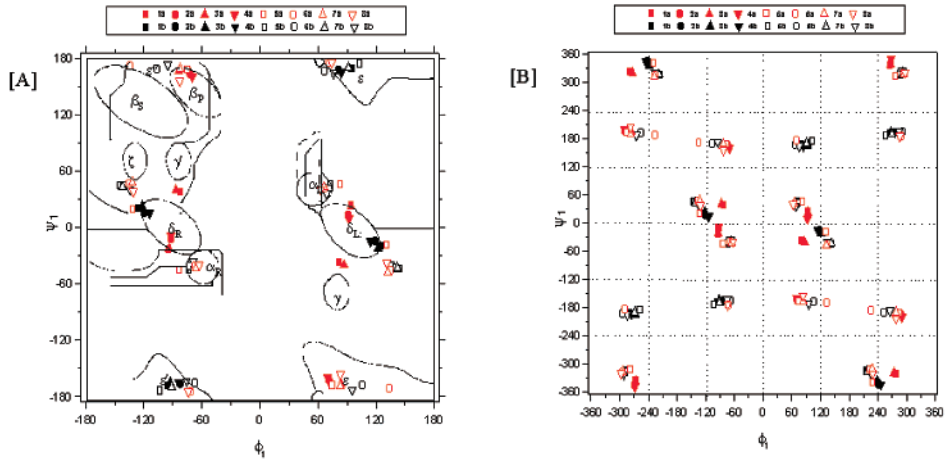

Figure 12. (A) Distribution of the backbone (ϕ_1, ψ_1) dihedral angles for the minimum energy conformations of *N*-methyl azapeptide derivatives **1**−**8** in the ϕ, ψ map. Allowed and partially allowed regions of native conformations and nomenclatures were taken from ref 30 (α_R, right-handed α-helix; α_L, mirror image of α-helix; β_S, region largely involved in β-sheet formation; β_P, regions associated with extended polyproline-like helix or β-sheet; γ and γ', γ and inverse-γ turns; δ_R, the bridge region; δ_L, mirror image of δ_R region; ϵ, extensive region with $\phi > 0$, $\varphi = \pm 180°$; ϵ' and ϵ'', mirror images of the two parts of the ϵ region; ζ, a region associated with residues preceding Pro). (B) Distribution of the backbone (ϕ_1, ψ_1) dihedral angles for the minima of **1**−**8** in two full cycles of the ϕ, ψ map.

from 347.6° to 355.1°. This demonstrates that a methyl group bound to the N2 enhances the planarity of the α-nitrogen atom, N2, and pyramidalization of the N2 atom in azapeptide derivatives including the azaGly residue can, as a result, decrease the amide conjugation between N2 and C3═O2 and increase the N2−C3 bond length. Thus, the N2−C3 bond is longer than the C2−N1 and N1−N2 bonds (see Supporting Information). These results show that the azapeptide derivatives including the azaGly residue lead to the pyramidalization of N2, while the N1 and N3 atoms are slightly pyramidalized. The average N2−C3 bond length in the azapeptide derivatives including azaAla is 1.408 Å, which is shorter than the average in the azapeptides including azaGly.

To understand the structural changes as a function of the addition of methyl groups on the N1, N2, and N3 atoms, the bond orders of *N*-methyl azapeptide derivatives **1**−**8** were calculated at the B3LYP/6-31G* level, and all of the bond orders for the C2−N1, N1−N2, N2−C3, and N2−C3 bonds in *N*-methyl azapeptide derivatives **1**−**8** were found to have single-bond character.[34] The average bond orders of the C2−N1, N1−N2, N2−C3, and C3−N3 bonds seem not to change upon the addition of methyl groups or with the orientation of the acetyl group. However, the rotational barriers of the N1−N2 and N2−C3 bonds are sufficiently large to restrict the free rotation, as shown in Table 3.

4. Conclusions

The distributions of the backbone (ϕ, ψ) angles of the minimum energy conformations for **1**−**8**, showing characteristic conformational preferences for *N*-methyl azapeptides, are superimposed in Figure 12, which represents the observed distribution of conformations in native protein and peptides.[27,30] The backbone dihedral angles of the minima for **1**−**8** were located mostly in allowed regions of native proteins and

(34) Reed, A. E.; Curtiss, L. A.; Weinhold, F. *Chem. Rev.* **1988**, *88*, 9.

peptides, which means that the minima of *N*-methyl azapeptides can be utilized to mimic native peptides or proteins. The *N*-methyl group substituent on the N1 or N2 atom in azapeptide derivatives does not perturb the backbone torsion angles significantly, whereas the additional *N*-methyl group on the N3 atom significantly changes the backbone torsion angles (see Figure 12). Also, the minima with a cis peptide bond in azapeptide derivatives **1**−**4** coupled to a primary amine are the lowest energy conformers, which is inconsistent with the fact that the native amino acids with a trans peptide bond are generally perferred over those with a cis peptide bond.[29,32] However, the minima with a trans peptide bond in azapeptide derivatives **5**−**7** coupled to a secondary amine are more stable than those with a cis peptide bond, except for **8**. Therefore, the *N*-methyl azapeptide derivatives can be utilized to determine the orientation of the amide bond. Furthermore, the helical structures were found as a local minimum conformation in compounds **5**−**8** coupled to a secondary amine. This shows that *N*-methyl azaamino acid derivatives can be utilized as α-helix initiators as well as various *β*-turn mimetics. The *N*-methyl group on the N2 atom of azapeptides including the azaAla residue leads to the planarity of the N2 atom, in comparison to the case of azapeptides including azaGly, which indicates that N2-methyl group can stabilize the urea-type amide resonance structure of azapeptide derivatives including the azaAla residue. This conformational information for azapeptide derivatives could play a critical role in designing useful molecules containing azaamino acids for drug discovery and peptide engineering.

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Supporting Information Available: Selected geometries of the B3LYP/6-31G* minimum energy conformations for model compounds **1**−**8**, selected Wiberg bond orders for **1**−**8**, and enlarged PESs of the critical section drawn with 1 kcal/mol increments for Figures 2A and 3A (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

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