

Conformational Analysis of a Cyclopropane Analogue of Phenylalanine with Two Geminal Phenyl Substituents

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Quantum mechanical methods have been used to investigate the intrinsic conformational preferences of 1-amino-2,2-diphenylcyclopropanecarboxylic acid (*c*₃Dip), a cyclopropane analogue of phenylalanine bearing two phenyl substituents on the same β -carbon. Geometries, energies, and frequencies were calculated on the *N*-acetyl-*N'*-methylamide derivative at the HF and B3LYP levels using the 6-31G(d), 6-311G(d), and 6-31+G(d,p) basis sets. Four minimum energy conformations were characterized: axial C₇, equatorial C₇, right-handed helix, and polyproline II. Analysis of the whole results, which are fully consistent with available experimental data, indicates that *c*₃Dip tends to promote γ -turn conformations.

Introduction

Among the quaternary α -amino acids used to constrain the flexibility of peptides, 1-aminocyclopropanecarboxylic acid (Ac₃c) shows particular conformational properties which have been well-characterized using both experimental¹ and theoretical² methodologies.

During the past years we have devoted a great deal of attention to investigate the conformational consequences produced by the incorporation of one or more phenyl groups to the cyclopropane ring of Ac₃c. The amino acids thus obtained can be regarded as phenylalanine analogues. We have shown experimentally³ and theoretically⁴ that their conformational propensities are influenced by the strained geometry of the three-membered system as well as by steric and electronic interactions between the rigidly held aromatic side chain(s) and the peptide backbone. Thus, studies on different stereoisomers of 1-amino-2-phenylcyclopropanecarboxylic acid (*c*₃Phe) showed that steric and electronic interactions between the rigidly held aromatic side chain and the main chain affect the conformational preferences to an extent that depends on the side chain orientation, i.e., on the *c*₃Phe stereochemistry.^{3a,b,4a} More recently, a tendency to promote folded structures in both solid state and solution was evidenced by the stereoisomers of 1-amino-2,3-diphenylcyclopropanecarboxylic acid with the phenyl substituents in a *trans* relative disposition (*c*₃diPhe).^{3c,4b}

In a very recent study,⁵ some of us incorporated a cyclopropane analogue of phenylalanine bearing two geminal phenyl substituents (1-amino-2,2-diphenylcyclopropanecarboxylic acid, *c*₃Dip) into a Pro-*c*₃Dip dipeptide. X-ray diffraction analysis showed that the (*S*)Pro-(*R*)*c*₃Dip stereoisomer adopts two consecutive γ -turns stabilized by intramolecular hydrogen bonds. This was the first observation of such a conformation among crystalline short linear peptides. Moreover, no other acyclic

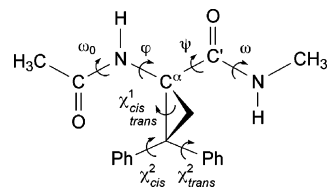


Figure 1. Structure of Ac-(*S*)*c*₃Dip-NHMe indicating the dihedral angles. The phenyl substituents are considered *cis* or *trans* according to their disposition relative to the *N*-terminus.

peptide containing a γ -folded proline residue was previously deposited in the Cambridge Structural Database. The ability of *c*₃Dip not only to adopt a γ -turn disposition but also to induce this structural motif in neighbor amino acids is expected to be of enormous interest in the design of bioactive peptides using conformationally restricted amino acids. Thus, recent studies on large peptides containing cyclopropane analogues of phenylalanine have demonstrated that selectively oriented side chains play a critical role in directing the backbone folding.⁶

Analysis of the intrinsic structural preferences of *c*₃Dip is essential to understand the potential of this amino acid to promote γ -turn conformations, the application of theoretical methods based on quantum mechanical calculations being specially appropriated for this task. In this work we examine the conformational preferences of the *N*-acetyl-*N'*-methylamide derivative of the *S* (or *L*) enantiomer of *c*₃Dip [Ac-(*S*)*c*₃Dip-NHMe (Figure 1)] using both *ab initio* and DFT methods.

Methods

The dipeptide Ac-(*S*)*c*₃Dip-NHMe retains the restrictions imposed in Ac-Ac₃c-NHMe by the cyclopropane ring, and additional conformational constraints arise from the presence of the two geminal phenyl substituents attached to it. Accordingly, the C₇ [(φ, ψ) = (−79°, 32°)], C₅ [(φ, ψ) = (180°, 180°)], and P_{II} [(φ, ψ) = (71°, −146°)] minimum energy conformations characterized for Ac-Ac₃c-NHMe at the HF/6-31G(d) level^{2,4a} were considered as good starting geometries for the conformational study of Ac-(*S*)*c*₃Dip-NHMe. However, due to the chiral

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TABLE 1: Dihedral Angles^a for the Conformational Energy Minima of Ac-(S)c₃Dip-NHMe at the HF/6-31G(d) and B3LYP/6-31G(d) Levels of Theory

| | ω_0 | j | y | χ^1_{cis} | χ^1_{trans} | χ^2_{cis} | χ^2_{trans} | w |
|----------------|------------|-------|--------|----------------|------------------|----------------|------------------|--------|
| HF/6-31G(d) | | | | | | | | |
| 1 | 171.7 | 75.5 | -43.3 | 1.9 | 147.7 | 77.1/-107.3 | 120.4/-63.3 | -177.2 |
| 2 | 171.6 | -72.4 | 119.7 | 3.6 | 148.9 | 74.9/-107.3 | 123.5/-60.2 | -173.6 |
| 3 | -168.2 | -79.9 | -24.3 | -0.2 | 143.1 | 77.9/-104.2 | 127.7/-56.3 | 173.0 |
| 4 | -160.4 | 63.8 | -173.9 | -1.0 | 144.4 | 74.4/-110.7 | 120.0/-64.8 | -179.7 |
| B3LYP/6-31G(d) | | | | | | | | |
| 1 | 171.7 | 72.1 | -42.3 | 1.8 | 146.9 | 75.3/-108.4 | 119.6/-63.9 | -177.2 |
| 2 | 179.0 | -77.2 | 93.5 | 4.1 | 149.4 | 75.1/-106.7 | 123.0/-61.5 | -177.3 |
| 3 | -169.9 | -80.2 | -20.4 | -0.7 | 141.8 | 78.9/-102.7 | 129.0/-55.5 | 173.4 |
| 4 | -158.7 | 63.7 | -173.6 | -1.7 | 142.8 | 73.6/-111.6 | 119.5/-64.1 | -179.3 |

^a In degrees; see Figure 1 for definition.

nature of the peptide under study, the mirror image structures were also built for the C₇ and P_{II} conformations of Ac-Ac₃C-NHMe. Accordingly, five backbone geometries were considered as starting points for geometry optimizations. Regarding the side chains, the values of χ^1 are fixed by the cyclopropane system, while three positions (*trans*, *gauche*⁺, and *gauche*⁻) were considered for χ^2_{cis} and χ^2_{trans} , which define the orientation of the phenyl rings. The dihedral angles related to the rotation of the amide bonds, ω_0 and ω , were arranged at 180° in all cases. Accordingly, 45 different structures, $5(\varphi, \psi) \times 3(\chi^2_{cis}) \times 3(\chi^2_{trans})$, were built, and those without severe steric conflicts between the phenyl side chains were taken as starting points in HF/6-31G(d)⁷ geometry optimizations. The resulting minimum energy conformations were fully reoptimized at the B3LYP/6-31G(d)⁸ level. Frequency analyses were performed in all cases to compute the conformational free energy differences in the gas phase at 298 K (ΔG^{gp}), using the standard statistical formulas to evaluate the zero-point vibrational energy (ZPVE) and both the thermal and entropic corrections.⁹ All the calculations were performed using the Gaussian 98 program.¹⁰

Results and Discussion

Only four minimum energy conformations were characterized for Ac-(S)c₃Dip-NHMe at the HF/6-31G(d) and B3LYP/6-31G(d) levels, their dihedral angles being listed in Table 1. This is an unexpected small number of minima taking into account that 45 conformations were taken as starting points in geometry optimizations. However, it should be noted that the relative arrangement of the phenyl side chains is dramatically restricted by both steric contacts and repulsive interactions between the electron densities of the π -systems. Thus, there is only one favorable arrangement for the phenyl groups, even although nine different dispositions were considered in the starting points. This feature is clearly reflected in Table 1, which shows that the dihedral angles χ^2_{cis} and χ^2_{trans} are very similar for all the backbone conformations, i.e., the disposition of the phenyl side chains is the same in the four minima. On the other hand, Table 2 lists the relative energy in the gas phase (ΔE^{gp}) and ΔG^{gp} for each minimum.

Conformer **1** corresponds to an axial C₇ (seven-membered hydrogen-bonded ring) or γ -turn conformation (Figure 2a) with hydrogen bonding parameters $d(\text{H}\cdots\text{O}) = 1.987 \text{ \AA}$ and $-\text{N}-\text{H}\cdots\text{O} = 148.4^\circ$ at the HF/6-31G(d) level [1.887 \AA and 151.9° at the B3LYP/6-31G(d) level]. Remarkably, the calculated (φ, ψ) angles are almost identical to those found⁵ in the crystalline structure of the aforementioned (S)Pro-(R)c₃Dip dipeptide [$(-72^\circ, 47^\circ)$, equivalent to $(72^\circ, -47^\circ)$ for (S)c₃Dip], as is the geometry of the hydrogen bond [$d(\text{H}\cdots\text{O}) = 1.97 \text{ \AA}$ and $-\text{N}-\text{H}\cdots\text{O} = 147^\circ$]. Of special relevance is the ability of theoretical calculations to predict the observed ψ -angle, which shows a

TABLE 2: Relative Energies^a (ΔE^{gp}) and Conformational Free Energies (ΔG^{gp} ; at 298 K) for the Minima of Ac-(S)c₃Dip-NHMe in the Gas Phase

| level ^b | | 1 | 2 | 3 | 4 |
|--------------------|-----------------|------------------|------------------|-----|-----|
| HF/6-31G(d) | ΔE^{gp} | 0.0 ^c | 0.6 | 1.7 | 3.3 |
| | ΔG^{gp} | 0.8 | 0.0 ^d | 1.1 | 4.1 |
| B3LYP/6-31G(d) | ΔE^{gp} | 0.0 ^e | 2.4 | 3.0 | 4.7 |
| | ΔG^{gp} | 0.0 ^f | 1.3 | 1.4 | 4.9 |
| HF/6-311G(d) | ΔE^{gp} | 0.0 ^g | 0.2 | 1.4 | 3.1 |
| | ΔG^{gp} | 1.0 | 0.0 ^h | 1.3 | 4.2 |
| B3LYP/6-311G(d) | ΔE^{gp} | 0.0 ⁱ | 1.8 | 2.7 | 4.4 |
| | ΔG^{gp} | 0.0 ^j | 1.7 | 2.0 | 4.3 |
| HF/6-31+G(d,p) | ΔE^{gp} | 0.0 ^k | 0.2 | 1.4 | 3.5 |
| | ΔE^{gp} | 0.0 ^l | 1.9 | 2.4 | 4.6 |

^a In kcal/mol. ^b Level of geometry optimization. ^c $E = -989.780112 \text{ au}$. ^d $G = -989.450729 \text{ au}$. ^e $E = -996.023933 \text{ au}$. ^f $G = -995.721063 \text{ au}$. ^g $E = -989.969145 \text{ au}$. ^h $G = -989.642679 \text{ au}$. ⁱ $E = -996.246563 \text{ au}$. ^j $G = -995.946476 \text{ au}$. ^k $E = -989.842998 \text{ au}$. ^l $E = -996.095520 \text{ au}$.

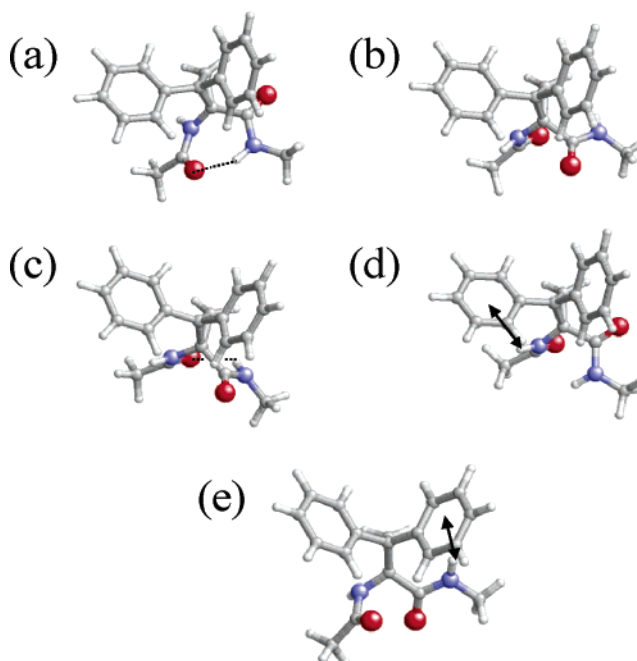


Figure 2. Minimum energy conformations of Ac-(S)c₃Dip-NHMe: (a) minimum **1** at the HF/6-31G(d) level; (b) minimum **2** at the HF/6-31G(d) level; (c) minimum **2** at the B3LYP/6-31G(d) level; (d) minimum **3** at the HF/6-31G(d) level; and (e) minimum **4** at the HF/6-31G(d) level. The parameters associated with the N-H \cdots O=C hydrogen bonds (dotted lines) and the N-H \cdots π interactions (lines with arrows) are given in the text.

significant deviation from the standard axial C₇ value ($\sim -70^\circ$). This tendency to adopt small ψ -angles has been related to hyperconjugation between the three-membered ring and the CO

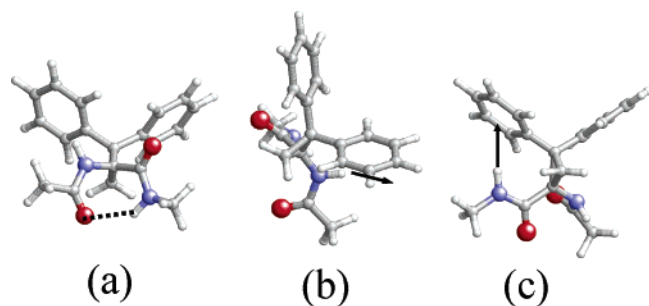


Figure 3. Selected details of the intramolecular interactions found for some minima of Ac-(S)₃cDip-NHMe: (a) intramolecular hydrogen bond (dashed line) of minimum **2** at the B3LYP/6-31G(d) level; (b) N–H··· π interaction (line with arrow) of minimum **3** at the HF/6-31G(d) level; and (c) N–H··· π interaction (line with arrow) of minimum **4** at the HF/6-31G(d) level.

substituent (maximal for $\psi = 0^\circ$) and was also observed in the C₇ minima of Ac₃c and the cyclopropane analogues of phenylalanine computed before.^{2,4} However, in these compounds, a clear preference to adopt conformations in the *bridge* [$(\varphi, \psi) = (80^\circ, 0^\circ)$, or the enantiomeric $(-80^\circ, 0^\circ)$] instead of the C₇ region has been established experimentally^{1,3} (note that the *bridge* region is attained through a further reduction in the ψ -angle of the C₇ minimum). In the case of c₃Dip, this deviation toward the *bridge* region has been observed⁵ in an (S)Pro-(S)₃cDip derivative [$(\varphi, \psi) = (69^\circ, -2^\circ)$], i.e., in only one out of the three crystalline structures reported to date^{5,8} for c₃Dip-containing peptides. This fact makes c₃Dip unique among cyclopropane amino acids, suggesting that it could be very useful to obtain stable building-block foldamers. Thus, there is a growing interest in the chemical manipulation of foldamer building blocks to adapt them for designed function under given environments, i.e., modification of protein modules for generating nanodevices.^{11,12} One strategy to enhance the stability of these nanomolecular architectures is through selected targeted replacement by nonproteogenic conformationally constrained amino acids.¹² The well-defined conformational properties of c₃Dip may help to exercise fine control over the re-engineered protein modules.

Although HF/6-31G(d) and B3LYP/6-31G(d) methods provide very similar geometries for **1**, important discrepancies are observed for minimum **2**. Thus, the HF/6-31G(d) structure can be identified as a polypeptide II (P_{II}) conformation (Figure 2b) with no intramolecular hydrogen bond [$d(\text{H}\cdots\text{O}) = 2.900 \text{ \AA}$ and $-\text{N}-\text{H}\cdots\text{O} = 108.6^\circ$], whereas the structure predicted at the B3LYP/6-31G(d) level corresponds to an equatorial C₇ conformation (Figure 2c) with hydrogen-bonding parameters $d(\text{H}\cdots\text{O}) = 2.211 \text{ \AA}$ and $-\text{N}-\text{H}\cdots\text{O} = 132.9^\circ$. This intramolecular interaction is displayed more clearly in Figure 3a. The difference between such structures must be attributed almost exclusively to the dihedral angle ψ , which is 26° smaller in the B3LYP/6-31G(d) minimum. Interestingly, the solid-state structure reported⁸ for *p*-BrBz-(S)₃cDip-NH^tPr with $(\varphi, \psi) = (-88^\circ, 152^\circ)$ corresponds to the P_{II} region of the conformational map, which is very rarely occupied by cyclopropane residues.¹ On the other hand, the characterization of a C₇ minimum at a high ψ -angle (93.5°), which disfavors hyperconjugation, is noteworthy. In fact, such a distortion of the γ -turn geometry was never observed in our previous computations on cyclopropane α -amino acids.^{2,4} It should be noted that small positive ψ -values are not allowed for the equatorial C₇ conformation of (S)₃cDip because they would place in high proximity the carbonyl oxygen and the *trans* phenyl group.

Inspection to the ΔE^{sp} values indicates that **1** is the most favored conformation, **2** being 0.6 and 2.4 kcal/mol unfavored at the HF/6-31G(d) and B3LYP/6-31G(d) levels, respectively. This relative energy order is preserved at the latter level of theory after transformation of ΔE^{sp} into ΔG^{sp} by adding the ZPVE and thermal and entropic corrections, although the difference with respect to **1** decreases by 1.1 kcal/mol. A similar reduction is obtained at the HF/6-31G(d) level (1.4 kcal/mol), but in this case an inversion in the relative stability order is observed. These results suggest that the ΔE^{sp} of minimum **2** is underestimated when electron correlation effects are neglected.

Minimum **3** corresponds to a right-handed 3₁₀- α -helical conformation (Figure 2d). This structure is stabilized by an interaction between the NH moiety of c₃Dip and the π -system of the *cis* phenyl ring, the N–H bond being more or less parallel to the plane of such phenyl. The parameters for this N–H··· π interaction, which is clearly depicted in Figure 3b, are $d_{\text{H}\cdots\text{Ph}} = 3.351 \text{ \AA}$ and $\theta = 23.9^\circ$ at the HF/6-31G(d) level [3.323 \AA and 24.7° at the B3LYP/6-31G(d) level], where $d_{\text{H}\cdots\text{Ph}}$ is the distance between the amide hydrogen and the center of the ring and θ is the angle between the N–H bond and the phenyl plane. Compared to the global minimum, **3** is unfavored by 1.7 and 3.0 kcal/mol at the HF/6-31G(d) and B3LYP/6-31G(d) levels, respectively. However, these values decrease by 0.6 and 1.6 kcal/mol, respectively, after adding the ZPVE, thermal, and entropic corrections to transform ΔE^{sp} into ΔG^{sp} .

Finally, minimum **4** is a P_{II} conformation with opposite handedness to that of **2** (Figure 2e). Interestingly, the geometries predicted using the HF and B3LYP methods are in excellent agreement, with no distortion toward the γ -turn being detected in this case. Figure 3c clearly shows that this semiextended structure is stabilized by an N–H··· π interaction between the methylamide NH and the *trans* phenyl substituent [$d_{\text{H}\cdots\text{Ph}} = 3.011 \text{ \AA}$ and $\theta = 46.5^\circ$ at the HF/6-31G(d) level; 2.955 \AA and 49.6° at the B3LYP/6-31G(d) level]. In this case the N–H bond and the phenyl ring are arranged more or less perpendicularly. Conformation **4** is the least favored in terms of both ΔE^{sp} and ΔG^{sp} .

Regarding the relative orientation of the phenyl planes, as explained above the dihedral angles χ^2_{cis} and χ^2_{trans} are very similar for the four minima characterized. Thus, the two aromatic rings adopt a hinge arrangement (Figure 2), which minimizes steric repulsions.

To check if the discrepancies found for **2** between HF and DFT calculations depend on the basis set, the four minima were reoptimized at the HF/6-311G(d), HF/6-31+G(d,p), B3LYP/6-311G(d), and B3LYP/6-31+G(d,p) levels. The resulting dihedral angles are listed in Table 3, while ΔE^{sp} and ΔG^{sp} values are included in Table 2. Unfortunately, frequency calculations were only possible at the HF/6-311G(d) and B3LYP/6-311G(d) levels and, therefore, ΔG^{sp} values have been calculated only at these levels of theory.

As can be seen, the results obtained with the 6-31G(d), 6-311G(d), and 6-31+G(d,p) basis sets are fully consistent within each method. Moreover, the differences predicted by HF and B3LYP methods for **2** are preserved when geometry optimizations are performed with the larger basis sets. Thus, the B3LYP/6-311G(d) and B3LYP/6-31+G(d,p) ψ -values are 22° and 16° smaller than those obtained at the HF/6-311G(d) and HF/6-31+G(d,p) levels, respectively, and allow the formation of a weak intramolecular hydrogen bond [$d(\text{H}\cdots\text{O}) = 2.295 \text{ \AA}$ and $-\text{N}-\text{H}\cdots\text{O} = 128.6^\circ$] and [$d(\text{H}\cdots\text{O}) = 2.584 \text{ \AA}$ and $-\text{N}-\text{H}\cdots\text{O} = 118.5^\circ$]. Furthermore, **2** becomes the minimum of lowest ΔG^{sp} at the HF/6-311G(d) level, even

TABLE 3: Dihedral Angles^a for the Conformational Energy Minima of Ac-(S)c₃Dip-NHMe at the HF/6-311G(d) and B3LYP/6-311G(d) Levels of Theory

| | ω_0 | j | y | χ^1_{cis} | χ^1_{trans} | χ^2_{cis} | χ^2_{trans} | w |
|-------------------|------------|-------|--------|----------------|------------------|----------------|------------------|--------|
| HF/6-311G(d) | | | | | | | | |
| 1 | 172.6 | 75.4 | -45.6 | 1.9 | 148.0 | 75.9/-108.4 | 118.4/-64.7 | -177.4 |
| 2 | 171.7 | -73.1 | 118.4 | 3.5 | 148.8 | 75.6/-106.6 | 123.4/-60.3 | -173.2 |
| 3 | -168.0 | -80.4 | -24.7 | -0.3 | 143.1 | 78.6/-103.4 | 127.5/-56.3 | 172.5 |
| 4 | -160.6 | 64.1 | -173.6 | -0.9 | 144.4 | 74.4/-110.7 | 118.7/-64.3 | -179.7 |
| HF/6-31+G(d,p) | | | | | | | | |
| 1 | 173.3 | 74.9 | -45.9 | 2.2 | 148.4 | 76.3/-107.8 | 117.7/-65.3 | -178.3 |
| 2 | 171.2 | -73.1 | 123.0 | 3.5 | 148.7 | 75.8/-106.4 | 123.2/-60.2 | -174.6 |
| 3 | -168.2 | -80.2 | -23.6 | -0.1 | 143.4 | 77.9/-104.2 | 125.5/-58.0 | 173.3 |
| 4 | -161.1 | 63.5 | -168.9 | -0.6 | 145.1 | 73.5/-111.3 | 116.4/-67.3 | -178.4 |
| B3LYP/6-311G(d) | | | | | | | | |
| 1 | 172.5 | 72.2 | -45.6 | 2.0 | 148.0 | 75.9/-110.0 | 118.4/-65.6 | -178.6 |
| 2 | 177.4 | -76.8 | 96.0 | 4.1 | 149.5 | 76.1/-105.7 | 124.2/-60.1 | -171.2 |
| 3 | -170.3 | -81.7 | -19.8 | -0.4 | 142.1 | 79.4/-102.2 | 128.8/-55.3 | 173.6 |
| 4 | -160.8 | 64.8 | -171.8 | -1.1 | 143.6 | 73.4/-111.7 | 119.2/-64.5 | -179.1 |
| B3LYP/6-31+G(d,p) | | | | | | | | |
| 1 | 174.9 | 71.5 | -45.5 | 2.4 | 148.0 | 75.5/-108.3 | 117.3/-65.8 | -179.3 |
| 2 | 176.8 | -75.6 | 107.1 | 3.9 | 149.0 | 76.0/-106.0 | 123.2/-60.8 | -172.7 |
| 3 | -170.8 | -80.7 | -20.3 | -0.3 | 142.5 | 78.7/-108.1 | 126.1/-57.7 | 173.6 |
| 4 | -162.9 | 63.8 | -162.3 | -0.2 | 145.1 | 72.3/-112.4 | 114.6/-69.5 | -178.2 |

^a In degrees; see Figure 1 for definition.

though the lowest ΔE^{sp} corresponds to **1**. Additionally, the ΔE^{sp} values predicted at the HF/6-311G(d) and HF/6-31+G(d,p) are almost identical, indicating that the addition of diffuse and polarization functions do not alter the results. This fact confirms that electron correlation effects are essential to provide reliable ΔE^{sp} values and, therefore, to discern between the relative stability of **1** and **2**. For minima **3** and **4**, the 6-31G(d), 6-311G(d), and 6-31+G(d,p) basis sets provide almost identical results.

As found previously for other cyclopropane analogues of phenylalanine,⁴ comparison of the present results with molecular mechanics calculations performed on a c₃Dip derivative¹³ is not satisfactory. In fact, methods based on classical mechanics were not able to predict either the relative stability or the geometry of the minima described here, and, thus, an α -helix was found¹³ as the most stable conformation for *p*-BrBz-(S)c₃Dip-NHⁱPr, with a geometry [$(\varphi, \psi) = (-69^\circ, -52^\circ)$] differing notably from that reported here (see the ψ value of minimum **3** in Tables 1 and 3). This deficiency is probably due to the unsuitability of the force-field parameters used in molecular mechanics calculations, which were directly transferred from proteinogenic α -amino acids.

The influence of the solvent on the relative stabilities of the four minima was estimated using the PCM model¹⁴ within the HF/6-31G(d) framework. Calculations were performed on the geometries optimized at the B3LYP/6-311G(d) level considering three different solvents: water, chloroform, and carbon tetrachloride. The conformational free energies in solution were estimated using the classical thermodynamics scheme, i.e., by adding the free energies of solvation to the gas-phase free energies.

Table 4 lists the conformational free energies in solution for conformations **1–4** in the three solvents. It is worth noting that **1** provides the most favorable interactions with the environment. Indeed, the relative stability of **1** increases progressively with the polarity of the solvent, the other three minima being unfavored by more than 4 kcal/mol in aqueous solution. This result is of particular relevance when compared with our previous calculations: for Ac₃c and the cyclopropane analogues of phenylalanine computed before, the γ -turn also appeared as the global minimum conformation but its relative stability decreased when the interaction with the solvent was considered.⁴

TABLE 4: Relative Conformational Free Energies^a in Solution for the Minima of Ac-(S)c₃Dip-NHMe

| solvent | 1 | 2 | 3 | 4 |
|-------------------|-----|-----|-----|-----|
| CCl ₄ | 0.0 | 2.2 | 2.4 | 4.7 |
| CHCl ₃ | 0.0 | 2.4 | 2.7 | 4.9 |
| H ₂ O | 0.0 | 4.6 | 4.8 | 4.3 |

^a In kcal/mol. The conformational free energies in solution were estimated by adding the free energies of solvation calculated using the PCM method to the relative free energies in the gas phase at the B3LYP/6-311G(d) level.

Summary

In summary, the calculations presented in this study indicate that c₃Dip exhibits the strongest tendency to adopt a γ -turn structure among the cyclopropane α -amino acids investigated to date, which is in good agreement with recently reported experimental data. The present work provides further evidence that the conformational properties of the cyclopropane analogues of phenylalanine can be modulated by changing the number and positions of the phenyl substituents. The results obtained in this study are currently used to generate new structures with nanoscale biological applications, i.e., nanoconstructs, replacing natural residues by c₃Dip at selected positions. More specifically, we are taking advantage of the potential of this constrained amino acid to promote turns enhancing the stability of the new nanoconstructs with respect to that obtained using natural amino acids. Furthermore, it should be noted that the hinge-like arrangement of the two phenyl rings provides a convenient means to form backbone–side chain (N–H $\cdots\pi$) and even side chain–side chain (π – π stacking) interactions with other residues, which may be of potential interest in both protein engineering and nanotechnology.

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Supporting Information Available: Coordinates of the minimum energy conformations characterized for Ac-(S)₃Dip-NHMe at different levels of theory. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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