

The Singular Gas-Phase Structure of 1-Aminocyclopropanecarboxylic Acid (Ac₃c)

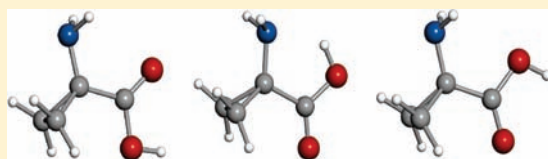
Ana I. Jiménez,[†] Vanesa Vaquero,[‡] Carlos Cabezas,[‡] Juan C. López,[‡] Carlos Cativiela,[†] and José L. Alonso^{*‡}

[†]Departamento de Química Orgánica, ISQCH, Universidad de Zaragoza—CSIC, 50009 Zaragoza, Spain

[‡]Grupo de Espectroscopía Molecular (GEM), Edificio Quifima, Laboratorios de Espectroscopia y Bioespectroscopia, Parque Científico Uva, Universidad de Valladolid, 45005 Valladolid, Spain

 Supporting Information

ABSTRACT: The natural nonproteinogenic α -amino acid 1-aminocyclopropanecarboxylic acid (Ac₃c) has been vaporized by laser ablation and studied in the gas phase by molecular-beam Fourier transform microwave spectroscopy. Comparison of the experimental rotational and ¹⁴N nuclear quadrupole coupling constants with the values predicted ab initio for these parameters has allowed the unambiguous identification of three Ac₃c conformers differing in the hydrogen bonding pattern. Two of them resemble those characterized before for the coded aliphatic α -amino acids. Remarkably, a third conformer predicted to be energetically accessible for all of these amino acids but never observed (the so-called “missing conformer”) has been found for Ac₃c, close in energy to the global minimum. This is the first time that such a conformer, stabilized by an N—H···O(H) hydrogen bond, is detected in the rotational spectrum of a gaseous α -amino acid with a nonpolar side chain. The conjugative interaction established between the cyclopropane ring and the adjacent carbonyl group seems to be responsible for the unique conformational properties exhibited by Ac₃c.



INTRODUCTION

α -Amino acids play an essential role in life. They not only participate in a plethora of physiological events, but also constitute the building blocks of peptides and proteins. The side chain diversity of the twenty genetically coded α -amino acids together with their intrinsic conformational flexibility make possible the wide variety of shapes and functions exhibited by proteins. The statistical analysis of crystallized proteins has yielded a great deal of information about the conformational preferences of α -amino acids when incorporated into peptide chains.¹ In comparison, much less is known about the conformational behavior of these building blocks considered as individual entities bearing the amino and carboxylic acid functionalities in the nonderivatized neutral form (NH₂, COOH). In condensed phases, amino acids are known to exist as ionized species. Moreover, in such environments, the ubiquitous intermolecular contacts may mask the structural properties inherent to the molecule under study. Therefore, only the gas phase provides adequate conditions to investigate the intrinsic structural properties of neutral amino acids.

Gas-phase studies are yet hampered by the difficulty to vaporize high-melting-point solids that, like amino acids, undergo thermal decomposition. Laser ablation (LA) has emerged as an attractive alternative to surmount the limitations associated to classical heating methods.² We have successfully applied this vaporization technique in combination with molecular-beam Fourier transform microwave (MB-FTMW) spectroscopy to delineate the conformational profile of different nonvolatile biologically relevant compounds, including several coded α -amino acids: alanine,³ valine,⁴

leucine,⁵ isoleucine,⁶ proline,⁷ serine,⁸ cysteine,⁹ threonine,¹⁰ and aspartic acid.¹¹ Our LA-MB-FTMW system¹² combines the advantages of laser-based vaporization and rotational spectroscopy in the environment of virtual isolation provided by a supersonic jet. An extremely high sensitivity and resolution is thus attained, as illustrated by the detection of some ¹⁵N and ¹³C species at natural abundance.^{7b,13,14} Furthermore, rotational spectroscopy is particularly suitable to study the conformational propensities of flexible molecules since it yields accurate structural information for each individual conformer present. Prior to our investigations, structural determinations of gaseous α -amino acids by microwave spectroscopy were limited to glycine¹⁵ and alanine.¹⁶

Although much work remains to be done in this field before the conformational landscape of all proteinogenic α -amino acids is known, the picture corresponding to those containing a nonfunctionalized aliphatic side chain is now complete. The coded aliphatic α -amino acids (including Gly) display a common structural profile governed by the hydrogen-bonding interactions established between the COOH and NH₂ moieties. Thus, all glycine,¹⁵ alanine,^{3,16} valine,⁴ leucine,⁵ and isoleucine⁶ have been shown to exist in the gas phase as a mixture of two conformers characterized by backbone geometries that are almost completely independent of the side-chain size (some variations are observed for glycine, vide infra). In all cases, the more populated form (I, Figure 1) exhibits a cis arrangement of the COOH group and a bifurcated hydrogen bond linking both amino hydrogen atoms to

Received: April 12, 2011

Published: June 01, 2011

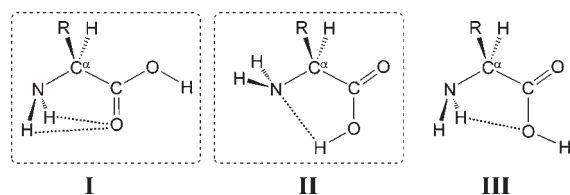


Figure 1. Different conformer types that can exist in α -amino acids according to the intramolecular hydrogen bonds established between the amino and carboxylic acid groups. Those detected by microwave spectroscopy for the coded aliphatic α -amino acids (I, II) are encircled.

the carbonyl oxygen ($\text{N}-\text{H}\cdots\text{O}=\text{C}$). The higher-energy conformer detected (II, Figure 1) is stabilized by a hydrogen bond between the nitrogen lone-pair electrons and the hydroxyl group ($\text{N}\cdots\text{H}-\text{O}$), which requires a trans-COOH unit.

A third type of hydrogen bond can be formed within the amino acid backbone, namely that with the hydroxyl oxygen acting as the hydrogen-bond acceptor ($\text{N}-\text{H}\cdots\text{O}-\text{H}$). In fact, conformers stabilized by this interaction (III, Figure 1) have been predicted to be energetically accessible for all coded aliphatic α -amino acids^{3–6,15,16} but never observed by microwave spectroscopy. Conformers I and III differ mainly in the orientation of the COOH group, which is rotated by ca. 180° around the $\text{C}_\alpha-\text{C}(\text{O})$ bond. Interconversion between them may therefore occur, provided that the energy barrier is low enough. Accordingly, the failure to detect type III conformers in the jet-cooled rotational spectra of coded aliphatic α -amino acids (indeed, of all α -amino acids with a nonpolar side chain investigated to date either genetically coded^{3–6,15,16} or not^{17,18}) has been attributed to conformational relaxation to the more stable I form during the cooling process associated to the supersonic-jet expansion.^{3–6,15e,16–19} An argument supporting this hypothesis is the observation of type III species in serine,⁸ cysteine,⁹ threonine,¹⁰ and aspartic acid¹¹ as a consequence of the hydrogen-bonding interactions established between the polar groups in the side chain (OH/SH/COOH) and those in the main chain. One may wonder whether the existence of a functionalized side chain able to form hydrogen bonds with the backbone is a prerequisite for the existence of conformer III in the cooled rotational spectrum of an α -amino acid. We hypothesized that α -amino acids lacking a polar side chain but exhibiting structural features leading to hindered rotation about the $\text{C}_\alpha-\text{C}(\text{O})$ bond would also present a III form amenable to detection. The nonproteinogenic 1-aminocyclopropanecarboxylic acid, known in the abbreviated form as Ac_3c , was selected to check the validity of this statement.

Ac_3c is a naturally occurring α -amino acid endowed with outstanding biological and pharmacological properties.²⁰ Moreover, when included into peptide chains, Ac_3c and its derivatives bearing additional substituents in the ring have shown remarkable conformational properties.²¹ Such a structural behavior presumably stems from the electron-donating capacity associated to the cyclopropane ring. The three-membered carbocycle has long been known for its ability to act as a π -electron donor and to conjugate with adjacent electron-acceptor centers such as multiple bonds, aromatic rings, and carbenium ions.²² The earliest evidence for this phenomenon was obtained almost one century ago from the reactivity²³ and UV spectra²⁴ of compounds containing a cyclopropane ring linked to a carbonyl group. The models for cyclopropane bonding proposed by Coulson and

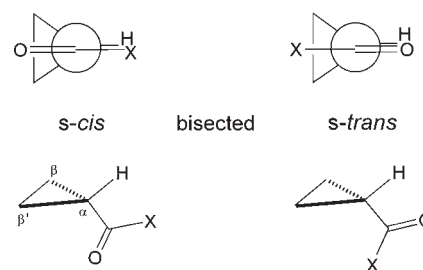


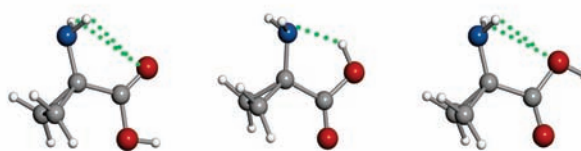
Figure 2. Schematic representation and Newman projections along the $\text{C}(\text{O})-\text{C}_\alpha$ bond illustrating the relative orientation of the cyclopropane ring and the carbonyl substituent in the *s-cis* and *s-trans* bisected conformations. The $\text{C}=\text{O}$ bond is eclipsed with the bisector of the cyclopropane $\text{C}_\beta-\text{C}_\alpha-\text{C}_{\beta'}$ angle in *s-cis* and with the $\text{C}_\alpha-\text{H}$ bond in *s-trans*.

Moffitt,^{25a} and Walsh^{25b} in the late 1940s stimulated the investigation of the factors that affect the conjugative ability of the three-membered ring, involving mainly carbonyl-derived π -acceptors (COX) and with special emphasis on the spatial requirements. Spectroscopic studies²⁶ as well as the analysis of X-ray structures²⁷ of a large number of carbonyl-substituted cyclopropanes provided conclusive evidence that this interaction is highly dependent on conformation. Electron transfer from the cyclopropane ring to the carbonyl substituent requires efficient overlapping of the interacting orbitals, and this has been shown to be maximal when the plane defined by the COX system bisects the three-membered cycle. This orientation corresponds to the so-called *s-cis* (synperiplanar) and *s-trans* (antiperiplanar) bisected conformations (Figure 2), which differ in a 180° rotation about the $\text{C}_\alpha-\text{C}(\text{O})$ bond. In the intermediate position, the two orbitals systems are orthogonal and overlapping is minimal.

According to electronic effects, the *s-cis* and *s-trans* bisected conformations should be equally favored for COX substituents. Numerous experimental and theoretical studies on the simplest carbonyl-substituted cyclopropane, cyclopropanecarboxaldehyde²⁸ ($\text{X} = \text{H}$), have shown that this compound exists in the two bisected forms, with the *s-trans* conformer being slightly more stable (energy differences generally not exceeding 100 cm^{-1} have been described). Comparison with the behavior exhibited by cyclopropanecarboxylic acid^{28h,i,29} ($\text{X} = \text{OH}$) and cyclopropyl methyl ketone^{28k,30} ($\text{X} = \text{Me}$) suggests that it is the steric demand of the X group that accounts for the predominance of one of the bisected forms. It has been established that the bulkier the X group, the higher the preference for the *s-cis* conformation. Thus, the *s-cis* population increases in the series $\text{COH} < \text{COOH} < \text{COCH}_3$. For the two latter substituents, the *s-trans* conformer is destabilized by about 350 and 700 cm^{-1} , respectively. In cyclopropanecarboxylic acid, both the *s-cis* and *s-trans* bisected forms present a *cis* arrangement of the COOH group. Most importantly, conjugation between the three-membered cycle and the carbonyl substituent not only restricts the orientations allowed to the substituent to the bisected conformations but also hinders rotation about the $\text{C}_\alpha-\text{C}(\text{O})$ bond. Torsional energy barriers in the neighborhood of 2000 cm^{-1} have been reported for the compounds mentioned^{28–30} and for other simple carbonyl-substituted cyclopropanes investigated.³¹ The aforementioned structural features made us consider Ac_3c a promising candidate for detection of the III conformer depicted in Figure 1.

Table 1. Molecular Properties of the Low-Energy Conformers of 1-Aminocyclopropanecarboxylic Acid (Ac₃c) Calculated *ab Initio*^a

Parameter ^b	I	II	III
A / MHz	3969.4	3940.2	4016.9
B / MHz	2657.7	2685.3	2623.4
C / MHz	1837.0	1844.8	1829.9
χ_{aa} / MHz	-4.85	-3.75	-4.93
χ_{bb} / MHz	3.03	1.93	3.07
χ_{cc} / MHz	1.82	1.82	1.86
$ \mu_a / D^c$	0.02	5.03	0.53
$ \mu_b / D$	1.19	3.61	1.32
$ \mu_c / D$	0.00	0.00	0.00
$ \mu_{total} / D$	1.18	6.19	1.42
$\Delta E_{MP2}^d / \text{cm}^{-1}$	0.0	303.4	108.1
$\Delta E_{MP2+ZPC}^e / \text{cm}^{-1}$	0.0	410.6	119.4



^a Optimized structures at the MP2/6-311++G(d,p) level of calculation. ^b A, B, and C are the rotational constants; χ_{aa} , χ_{bb} , and χ_{cc} are the ¹⁴N nuclear quadrupole coupling constants (diagonal elements of the quadrupole coupling tensor); μ_a , μ_b , and μ_c are the electric dipole moment components. ^c 1 D $\approx 3.3356 \times 10^{-30}$ C m. ^d Uncorrected for zero-point vibrational energies. ^e Relative electronic energies including B3LYP/6-311++G(d,p) harmonic zero-point vibrational energies.

EXPERIMENTAL SECTION

The LA-MB-FTMW spectrometer has been described in detail elsewhere.^{10,12b} A sample of Ac₃c (mp 231 °C) was obtained as previously reported.³² This compound has recently become available from commercial sources. Finely powdered Ac₃c was mixed with minimum quantities of a commercial binder and pressed to form cylindrical target rods. The rods are accommodated in a special pulsed injection nozzle located at the backside of one of the mirrors forming the Fabry-Pérot resonator. The molecules are vaporized with 532 nm green pulses (~ 10 ns) of a Q-switched Nd:YAG laser focused to give irradiances ca. 10^8 W cm⁻². The ablation products are seeded in an inert gas stream and entrained into a vacuum chamber, where they expand supersonically to form a molecular beam between the mirrors of the Fabry-Pérot resonator. Neon at stagnation pressures ca. 5 bar was used as the carrier gas with nozzle openings of 0.3–0.7 ms. A microwave pulse of 0.2–0.4 μ s (1–30 mW) is then applied to the center of the resonator through a coaxial antenna to cause the macroscopic polarization of the molecules in the beam. The emission signal produced upon molecular relaxation (free-induction-decay, FID) is recorded in the time domain and subsequently Fourier-transformed to the frequency domain to obtain the rotational spectrum. Due to the collinear arrangement of the supersonic jet and the resonator axis, each line of the spectrum appears as a Doppler doublet. The frequencies of the rotational transitions are calculated as the arithmetic mean of the two Doppler components. The accuracy of the frequency measurements is estimated to be better than 3 kHz.

RESULTS AND DISCUSSION

Theoretical calculations have been used to identify the most stable structures of Ac₃c and to predict their molecular properties relevant for the analysis of the rotational spectra. The three conformations in Figure 1, differing in the orientation of the amino and carboxylic acid groups and the hydrogen-bonding

interactions connecting them, were considered as starting geometries. Full geometry optimizations were performed using second-order Møller–Plesset perturbation theory in the frozen core approximation with a Pople's 6-311++G(d,p) basis set.³³ This level of theory has been proven to satisfactorily predict the molecular properties of amino acids in our previous studies. The spectroscopic parameters calculated for the three low-energy conformers of Ac₃c are listed in Table 1, where their geometries and relative energies are also shown. The Cartesian coordinates of all atoms in the principal inertial axis system are given in the Supporting Information. Calculation of relative stabilities was improved by including the zero-point vibrational contributions derived using the B3LYP functional in the harmonic approximation. Conformer I was predicted to be the global minimum, with II and III exhibiting relative energies of 411 and 119 cm⁻¹, respectively. Accordingly, the gaseous equilibrium mixture of Ac₃c should contain significant amounts of all three conformers. Moreover, all of them should be amenable to detection, as non-negligible values have been predicted for their electric dipole moment components along at least one inertial axis (Table 1). However, it should be born in mind that similar considerations regarding energy accessibility and dipole moment values applied to the I, II, and III conformers characterized theoretically for the coded aliphatic α -amino acids studied before, yet the latter form eluded experimental detection in all cases.^{3–6,15,16} The same holds true for the α -amino acids bearing a nonpolar side chain of nonproteinogenic nature investigated to date.^{17,18}

The conformational searches in the microwave spectra were next performed using the calculated spectroscopic parameters in Table 1 as a guide. In particular, the rotational constants (A, B, C), which depend on the mass distribution and geometry, and the ¹⁴N nuclear quadrupole coupling constants (diagonal elements χ_{aa} , χ_{bb} , χ_{cc}), which reflect the chemical environment around

Table 2. Experimental Rotational Parameters of the Three Conformers Observed for 1-Aminocyclopropanecarboxylic Acid (Ac₃c)

parameter ^a	I	II	III
A/MHz	3973.82253(93) ^b	3951.76469(89)	4026.9318(12)
B/MHz	2656.29291(47)	2678.63298(43)	2618.61756(57)
C/MHz	1838.42702(39)	1844.94862(33)	1831.11726(53)
Δ _J /kHz	0.234(17)	0.245(13)	0.241(22)
P _c /uÅ ²	21.2684 (5)	21.3158 (5)	21.2497(7)
χ _{aa} /MHz	-4.4849(17)	-3.4242(13)	-4.6145(21)
χ _{bb} /MHz	2.9016(27)	1.8420(22)	2.9396(36)
χ _{cc} /MHz	1.5832(27)	1.5822(22)	1.6748(36)
σ/kHz	1.7	1.7	2.0
N	24	36	24

^a A, B, and C are the rotational constants; Δ_J is the quartic centrifugal distortion constant; P_c = 1/2 (I_a + I_b - I_c) is the planar moment of inertia, conversion factor: 505379.1 MHz uÅ²; χ_{aa}, χ_{bb}, and χ_{cc} are the ¹⁴N nuclear quadrupole coupling constants (diagonal elements of the quadrupole coupling tensor); σ is the rms deviation of the fit; N is the number of fitted transitions. ^b Standard error in parentheses in units of the last digit.

the nitrogen nucleus, are invaluable tools for conformer identification.¹⁰ Additionally, the microwave power required to optimally polarize a certain conformer and the intensity of the transition lines observed should be consistent with the calculated magnitude of the electric dipole moment components along the principal inertial axes (μ_a, μ_b, μ_c).

According to the data in Table 1, rotational spectra with $J \leq 8$ in the 4–12 GHz region were expected for the three Ac₃c conformers. The search started on the basis of the predictions made for the II form. The large value of its μ_a electric dipole moment component (Table 1) should result in an intense rotational spectrum. Initial surveys soon identified the characteristic μ_a-type R-branch 2_{0,2}←1_{0,1} and 2_{1,2}←1_{1,1} rotational transitions. These measurements were extended to other μ_a- and μ_b-type R-branch transitions, which led to the assignment of a first rotamer for Ac₃c. No transitions obeying μ_c-type selection rules could be observed, indicating a very small or zero electric dipole moment component along the *c* axis. The absence of μ_c-type transitions and the low microwave power needed for optimal polarization of the μ_a- and μ_b-type transitions pointed to the detection of conformer II. Apart from the instrumental Doppler effect doubling, all measured transitions were split into several close hyperfine components attributable to the nuclear quadrupole coupling interaction originated by the presence of a ¹⁴N (*I* = 1) nucleus. This confirmed that the spectrum was generated by a molecule containing a single nitrogen atom. All measured transitions were fitted using the Hamiltonian $H = H_R^{(A)} + H_Q$, where $H_R^{(A)}$ represents the Watson's *A*-reduced semirigid rotor Hamiltonian³⁴ and H_Q is the nuclear quadrupole coupling interaction term.³⁵ The Hamiltonian matrix was directly diagonalized³⁶ to yield the spectroscopic parameters listed in the second column of Table 2. The experimental rotational constants obtained matched nicely with those predicted ab initio for conformer II (Table 1), thus providing further support for its identification. However, definitive confirmation of the identity of this conformer came from the agreement between the measured and calculated ¹⁴N nuclear quadrupole coupling constants (Tables 1 and 2). The latter parameters are extremely sensitive

to the orientation of the amino group with reference to the principal inertial axes and provide independent guidance for conformer discrimination.

Once the analysis of conformer II was concluded, the spectrum was surveyed for additional species. After long scans, a set of μ_b-type R-branch transitions of a second rotamer exhibiting the expected ¹⁴N nuclear quadrupole coupling hyperfine patterns was readily assigned. No transitions following μ_a- or μ_c-type selection rules could be detected, suggesting negligible dipole moment components in these directions, as predicted for the type I form (Table 1). The analysis of the rotational transitions measured was performed using the same Hamiltonian and provided the parameters given in the first column of Table 2. Comparison of the experimental rotational and nuclear quadrupole coupling constants obtained for this rotamer with the calculated values in Table 1 allowed the unequivocal identification of conformer I.

In our previous studies of α-amino acids containing a nonpolar side chain,^{3–6,17,18} no transitions attributable to other conformers remained in the spectra after the assignment of rotamers of the I and II types. We were pleased to observe that this was not the case in the present work. The search was pursued since a significant number of transitions, presumably belonging to conformer III, still remained unassigned in the rotational spectrum of Ac₃c. After discarding the lines corresponding to the I and II forms, μ_a- and μ_b-type R-branch transitions of a third rotamer were assigned. The observation of ¹⁴N nuclear quadrupole coupling hyperfine structures confirmed the presence of a nitrogen-containing species. Analysis of the rotational transitions yielded the parameters listed in the third column of Table 2. The good agreement between these data and the predicted rotational and ¹⁴N nuclear quadrupole coupling constants in Table 1 provided the definitive proof to identify the new rotamer as III. It is worth noting that unambiguous discrimination between the I and III species could not have been done on the basis of the rotational and ¹⁴N nuclear quadrupole coupling constants alone, since both conformers present a similar distribution of the heavy atoms and an identical orientation of the amino moiety. Accordingly, the two forms should exhibit close values for the rotational and nuclear quadrupole coupling constants, as in fact was predicted ab initio and found experimentally (Tables 1 and 2). However, the different magnitude of the dipole moment component along the *a* axis calculated for the I and III conformers is in accordance with the detection of μ_a-type transitions only for the latter and unequivocally verifies the assignment made.

No transitions imputable to other conformers were left unassigned in the rotational spectrum of Ac₃c. A full list of the transition frequencies measured for the three rotamers is provided as Supporting Information.

Therefore, the so far called “missing conformer” for its ability to elude detection in apparent contradiction to the theoretical predictions, has finally been observed in the jet-cooled rotational spectrum of an α-amino acid containing a nonpolar side chain. As mentioned above, the type III conformer was predicted to be energetically accessible for all aliphatic α-amino acids investigated but never observed.^{3–6,15–17} Its elusiveness has been attributed to conformational relaxation to the more stable I form during the supersonic expansion.^{3–6,15e,16–19} Thus, it has been proposed that the III conformer is actually populated in the initial equilibrium mixture formed but disappears during the strong cooling associated to the adiabatic expansion. Such a cooling process is expected to bring all species initially present in the

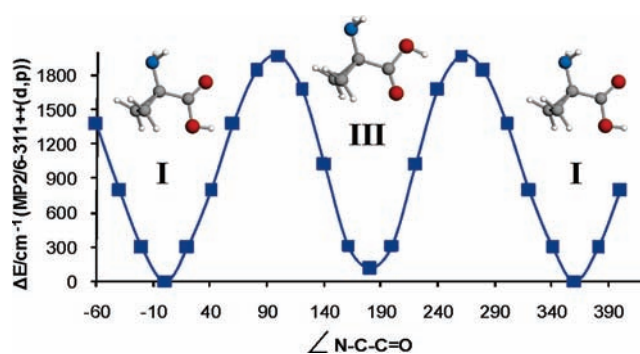


Figure 3. Profile of the interconversion between the I and III conformers of Ac_3c through rotation about the $\text{C}_\alpha\text{-C}(\text{O})$ bond calculated at the MP2/6-311++G(d,p) level.

mixture to their ground vibrational states but some local minima may concomitantly collapse to lower-energy forms if they are separated by affordable barriers (a threshold value of 400 cm^{-1} has been proposed for systems involving one degree of freedom at room temperature³⁷). Other plausible explanations for the failure to detect III are a low population in the beam or a very small dipole moment or a combination of both factors making this conformer fall below the detection limit. However, this is rather unlikely considering the theoretical predictions and the extremely high sensitivity of our instrument. The possibility that computations greatly overestimate the stability of this conformer type cannot be completely discarded but seems little probable. The fact that conformers of the III category predicted to be thermally accessible have actually been detected^{8–11} in α -amino acids bearing a polar side-chain able to establish hydrogen bonds with the backbone provides an argument in favor of conformational relaxation. The observation of III in Ac_3c further supports the existence of this species in the pre-expansion equilibrium mixture of other aliphatic α -amino acids. Furthermore, it verifies our initial hypothesis that type III conformers can be detected in the absence of polar substituents.

The observation of the III conformer in Ac_3c is highly remarkable but not completely unexpected according to the considerations in the Introduction section. Relaxation from the III to the I form involves rotation around the $\text{C}_\alpha\text{-C}(\text{O})$ bond and this conformational change should be highly restricted in Ac_3c due to conjugation between the cyclopropane ring and the carboxylic acid. We have studied theoretically the interconversion between the I and III forms of Ac_3c at the MP2/6-311++G(d,p) level of theory. A barrier around 2000 cm^{-1} , centered at the position where the interacting cyclopropane and carbonyl orbitals are perpendicular, has been encountered (Figure 3). This value is in line with those reported for other carbonyl-substituted cyclopropanes^{28–31} and high enough to preclude interconversion between the I and III conformers of Ac_3c . In comparison, energy barriers below 400 cm^{-1} have been calculated for the $\text{C}_\alpha\text{-C}(\text{O})$ torsion associated to the III \rightarrow I conversion in aliphatic α -amino acids previously studied.^{3,17,19}

The detection of the III Ac_3c conformer also proves that hampering relaxation from the III to the I form may be achieved through side-chain to main-chain interactions different from hydrogen bonding. Prior to the study of Ac_3c , the conformational landscape observed for all of the α -amino acids investigated, independently of their coded or noncoded character or their side-chain nature, was found to be the consequence of the hydrogen

bonds established between the polar groups in the molecule. This is the first occasion on which interactions other than hydrogen bonding are shown to be responsible for the stabilization of a particular conformer type in an α -amino acid. It is worth mentioning that in our studies of β -alanine³⁸ and γ -aminobutyric acid,³⁹ i.e., the glycine homologues bearing one or two additional methylene groups in the backbone respectively, some conformers were found to be stabilized by an interaction between the amino and carbonyl groups different from hydrogen bonding, namely of the $n \rightarrow \pi^*$ type. Therefore, the analysis of Ac_3c has allowed the characterization of an intramolecular force able to govern the conformational propensities of the backbone not detected before in the ensemble of amino acids (of the α , β , or γ type) studied in the gas phase.

Notably enough, the presence of the cyclopropane moiety is more effective in hindering the III \rightarrow I relaxation process than hydrogen bonding has proven to be in the polar α -amino acids investigated (serine,⁸ cysteine,⁹ threonine,¹⁰ and aspartic acid¹¹). In fact, the number of type III conformers predicted to be thermally accessible for such compounds is superior to that observed (note that the presence of a polar side chain increases the number of energy minima for a given backbone arrangement and therefore various conformers of the I, II, and III categories have been characterized as low-energy minima for these amino acids). This suggests that precluding rotation about the $\text{C}_\alpha\text{-C}(\text{O})$ bond through the intramolecular hydrogen-bonding network generated by the presence of a polar side chain is effective in a limited number of cases. Moreover, the energy barriers hindering the III \rightarrow I relaxation in such positive cases^{10,11} were estimated to be around 1000 cm^{-1} , that is, substantially inferior to that calculated for Ac_3c (Figure 3). It should be noted that partial relaxation has been observed³⁸ between conformers of β -alanine separated by a barrier of 1000 cm^{-1} . Accordingly, conjugation between the cyclopropane ring and the carboxylic acid is the most effective interaction in hindering the $\text{C}_\alpha\text{-C}(\text{O})$ torsion, and therefore the III \rightarrow I conformational relaxation, among those detected to date in α -amino acids. Indeed, for the very first time in our investigations, the number of conformers observed in the jet-cooled rotational spectrum of an amino acid does comprise all of the energetically accessible forms corresponding to the equilibrium distribution predicted by ab initio computations.

The relative abundances of the Ac_3c conformers detected in the supersonic jet have been obtained considering the intensities of four μ_b -type R-branch transitions measured for all three ($2_{0,2} \leftarrow 1_{1,1}$, $2_{1,2} \leftarrow 1_{0,1}$, $3_{1,3} \leftarrow 2_{0,2}$, and $3_{0,3} \leftarrow 2_{1,2}$). The intensities of these lines should be proportional to the concentration of each species in the beam scaled by the corresponding μ_b electric dipole moment component (Table 1). On this basis, the approximate population ratio has been estimated to be I:II:III = 6:1:3, which is in good agreement with the relative energies calculated ab initio (Table 1). Furthermore, no variation in the line intensities was observed when argon was used instead of neon as the carrier gas. This result reinforces the idea that no conformer interconversion occurred since heavier gases are known to favor this process.³⁷ It should be emphasized that Ac_3c is the only amino acid investigated so far by these methods for which conformational relaxation can be discarded.

The conjugation established between the three-membered cycle and the adjacent carbonyl group also influences the geometry of the Ac_3c conformers. For the three species detected, there is an excellent agreement between the measured and calculated

rotational constants. Thus, scale factors ranging from 0.997 to 1.003 bring the predicted parameters in Table 1 into coincidence with the experimental ones in Table 2, which means a maximum deviation of 0.3%. This indicates that the actual geometries of the Ac_3c conformers very close resemble those calculated *ab initio* (Table 1). All of them present a C_s symmetry, with the backbone heavy atoms lying on the plane that bisects the cyclopropane ring. The experimental value obtained for the planar moment of inertia P_c (Table 2), which reflects the mass contributions out of the *ab* inertial plane, is consistent with that expected for a C_s symmetry in all three conformers, with two equivalent out-of-plane cyclopropane methylene groups and amino hydrogens. The nonobservation of μ_c -type transitions is also consistent with a C_s symmetry, for which a zero electric dipole moment component along the *c* axis is expected (Table 1).

Although the geometry of the Ac_3c conformers could be regarded as a direct consequence of the nonchiral nature of the molecule, the structure of rotamer III deserves particular comment. Its C_s symmetry cannot be attributed to the symmetrical character of the cyclopropane side chain since the C_s symmetry exhibited by the I and II forms in glycine is not maintained in the type III conformer. Thus, the glycine I and II conformers have been shown to present a planar heavy-atom skeleton, whereas the III form is predicted to be nonsymmetrical, with the $\text{N}-\text{C}_\alpha-\text{C}=\text{O}$ dihedral angle deviating significantly from planarity (a $\text{C}_\alpha-\text{C}(\text{O})$ torsion near 30° has been calculated⁴⁰). A similar pattern applies to the chiral aliphatic α -amino acids studied,^{3-6,17} although in these cases the I and II forms are characterized by a quasi-planar heavy-atom backbone (deviations slightly above 10° have been observed for alanine³) and a higher deformation from planarity is predicted for III. It ensues that the geometry of conformers I and II in glycine is related to the symmetrically substituted α carbon but clearly another factor operates in the III species that is responsible for the distortion from the planar framework.

The amino group acts as a hydrogen-bond donor in both the I and III minima (Figure 1). In the former case, a bifurcated interaction is established with the carbonyl oxygen whereas only one amino hydrogen is linked to the hydroxyl oxygen in III. The different orientation of the lone-pair orbitals of each oxygen type could be at the basis of the different relative arrangement adopted by the hydrogen-bond donor and acceptor in these species. If such is the case, then optimization of the hydrogen-bond interaction should be responsible for the different geometry regarding backbone planarity displayed by the I and III conformers in the aliphatic α -amino acids so far studied. It appears that the III Ac_3c conformer does not follow this trend and this distinct behavior may be ascribed again to the conjugative ability of the cyclopropane ring. Optimization of the hydrogen bond in III apparently requires torsion around the $\text{C}_\alpha-\text{C}(\text{O})$ angle which, in the case of Ac_3c , implies the deviation of the carbonyl group from the preferred bisected conformation (Figure 2) and a concomitant less efficient overlap of the cyclopropane and carbonyl interacting orbitals at a high energy cost (as much as 500 cm^{-1} for a 30° torsion; Figure 3). It seems that the energy penalty associated to deviation from the bisected position is superior to that derived from a less favorable hydrogen-bond geometry and, as a consequence, the III conformer in Ac_3c retains the planar backbone geometry and the symmetrical bifurcated hydrogen bond encountered in the I form. The C_s symmetry of the III species in Ac_3c can thus be viewed as a further proof of the conjugative interaction established between the three-membered cycle and the contiguous carbonyl group.

From the above discussion, it becomes apparent that not only the detection but also the shape of the III Ac_3c conformer is particular. The cyclopropane unit clearly exerts a large influence on the structural propensities of Ac_3c , in contrast to the aliphatic side chains previously studied. Ac_3c is indeed the first α -amino acid investigated in the gas phase in which a nonpolar side chain is seen to be a major determinant of the backbone structure.

At this stage, one may wonder about the role played by the amino group in the conformational propensities observed for Ac_3c apart from its participation in hydrogen bonding. The question arises as to whether any specific interaction between the amino group and the cyclopropane ring could be responsible, to a certain extent, for the conformational behavior exhibited by Ac_3c . In contrast to the well-established capacity of the three-membered carbocycle to donate electron density to π -acceptors,^{22,26,27} its interaction with electron-rich substituents like the amino group is more complex and not completely understood.^{22b,27,41} Cyclopropylamine has been shown to exist as a mixture of two conformers.^{41,42} The lowest-energy form is characterized by a symmetrical structure, in which the two $\text{N}-\text{H}$ bonds are staggered with respect to the cyclopropane bonds and the nitrogen lone-pair orbital points toward the cycle.^{41,42} This arrangement corresponds to that encountered in the I and III conformers of Ac_3c . The minor cyclopropylamine conformer is about 700 cm^{-1} higher in energy and presents the amino group rotated by ca. 130° with respect to the preferred orientation.^{42b,c,g} Although conjugative interactions were initially invoked^{42a} to explain the structure of the dominant conformer, the finding that the energy barriers associated to torsion around the $\text{N}-\text{C}$ angle are similar in cyclopropylamine and isopropylamine^{42c} provided a solid argument against the existence of π -electron transfer from the amino to the cyclopropane bonds. The structure of cyclopropylamine is currently assumed to be mainly the consequence of minimization of the repulsive interactions between the electron-rich regions in the molecule.

While the orientation of the amino group in the I and III conformers of Ac_3c corresponds to that predominating in cyclopropylamine, that in II is not among those characterized as energy minima for the parent compound. It seems, therefore, that the amino group in Ac_3c is able to adopt conformations that are not favorable in the absence of the neighboring carboxylic acid. In comparison, the marked propensity of cyclopropanecarboxylic acid to adopt a bisected conformation (Figure 2) is strictly maintained in Ac_3c , even when, as seen for the III conformer, this implies a less favorable geometry for the intramolecular hydrogen bond. Thus, the conformational propensities of cyclopropanecarboxylic acid prevail over those of cyclopropylamine. In fact, the role played by the amino group in Ac_3c does not seem to differ much from that observed in other α -amino acids and no evidence has been found for it being responsible for the structural singularities observed for Ac_3c . Accordingly, such structural features should be ascribed exclusively to the cyclopropane-carbonyl conjugation. The fact that the II Ac_3c conformer is higher in energy than I and III may be due to the less favorable orientation of the amino group with respect to the cyclopropane ring discussed above but also to the trans arrangement of the COOH group (Figure 1), which is known to be much less favorable than the cis state.⁴³

CONCLUSIONS

Three different conformers have been detected in the rotational spectrum of 1-aminocyclopropanecarboxylic acid (Ac_3c).

Their structure has been unambiguously assigned by comparing the values observed experimentally for the rotational and quadrupole coupling constants with those provided by *ab initio* calculations. Their relative abundances in the cooled jet are coincident with those predicted theoretically, suggesting that the conformer distribution has been preserved during the expansion.

The side chain plays a primary role in driving the conformational properties of Ac₃c. This is in sharp contrast with the behavior exhibited by the aliphatic α -amino acids studied before by microwave spectroscopy. Thus, a type III conformer stabilized by an N–H \cdots O–H hydrogen bond has been observed for the first time among α -amino acids devoid of a functionalized side chain. Moreover, this Ac₃c conformer is almost isoenergetic with the global minimum (I) and interconversion between them is hindered by an extremely high energy barrier. The fact that the III conformer detected presents C_s symmetry is also striking. The conjugative interaction established between the cyclopropane ring and the adjacent carbonyl group appears to be at the basis of the particular structural features of Ac₃c.

This is the first α -tetrasubstituted α -amino acid characterized in the gas phase by rotational spectroscopy. However, the distinct conformational behavior observed for Ac₃c should be ascribed to the conjugative ability of the cyclopropane ring rather than to its α -tetrasubstituted character, although a simultaneous influence of both factors cannot be discarded. Studies to discern to which extent α -tetrasubstitution may affect the conformational propensities of α -amino acids are in progress.

Although the conformational preferences determined for gaseous Ac₃c in this work are not expected to be strictly maintained when incorporated into a peptide chain, the results obtained help understand the particular structural behavior observed for peptides containing cyclopropane α -amino acids. They also constitute a valuable proof that nonproteinogenic amino acids may display conformational features different from that characterizing the coded α -amino acids and therefore be useful in the design of peptides with singular structural properties.

■ ASSOCIATED CONTENT

S Supporting Information. Complete list of transition frequencies measured, Cartesian coordinates for the three Ac₃c conformers, and complete ref 33. This material is available free of charge via the Internet at <http://pubs.acs.org>.

■ AUTHOR INFORMATION

Corresponding Author

jlalonso@qf.uva.es

■ ACKNOWLEDGMENT

This research was funded by the Ministerio de Ciencia e Innovación (Grant Nos. CTQ2006-05981/BQU, CTQ2010-17436, and Consolider-Ingenio 2010 CSD2009-00038), Junta de Castilla y León (Grant No. VA070A08), and Gobierno de Aragón (research group E40). V.V. and C.C. thank the Ministerio de Ciencia e Innovación for FPU and FPI grants, respectively.

■ REFERENCES

- Chakrabarti, P.; Pal, D. *Prog. Biophys. Mol. Biol.* **2001**, *76*, 1–102.
- Levis, R. J. *Annu. Rev. Phys. Chem.* **1994**, *45*, 483–518.
- Blanco, S.; Lesarri, A.; López, J. C.; Alonso, J. L. *J. Am. Chem. Soc.* **2004**, *126*, 11675–11683.
- Lesarri, A.; Cocinero, E. J.; López, J. C.; Alonso, J. L. *Angew. Chem., Int. Ed.* **2004**, *43*, 605–610.
- Cocinero, E. J.; Lesarri, A.; Grabow, J.-U.; López, J. C.; Alonso, J. L. *Chem. Phys. Chem.* **2007**, *8*, 599–604.
- Lesarri, A.; Sánchez, R.; Cocinero, E. J.; López, J. C.; Alonso, J. L. *J. Am. Chem. Soc.* **2005**, *127*, 12952–12956.
- (a) Lesarri, A.; Mata, S.; Cocinero, E. J.; Blanco, S.; López, J. C.; Alonso, J. L. *Angew. Chem., Int. Ed.* **2002**, *41*, 4673–4676. (b) Mata, S.; Vaquero, V.; Cabezas, C.; Peña, I.; Pérez, C.; López, J. C.; Alonso, J. L. *Phys. Chem. Chem. Phys.* **2009**, *11*, 4141–4144.
- Blanco, S.; Sanz, M. E.; López, J. C.; Alonso, J. L. *Proc. Natl. Acad. Sci. U.S.A.* **2007**, *104*, 20183–20188.
- Sanz, M. E.; Blanco, S.; López, J. C.; Alonso, J. L. *Angew. Chem., Int. Ed.* **2008**, *47*, 6216–6220.
- Alonso, J. L.; Pérez, C.; Sanz, M. E.; López, J. C.; Blanco, S. *Phys. Chem. Chem. Phys.* **2009**, *11*, 617–627.
- Sanz, M. E.; López, J. C.; Alonso, J. L. *Phys. Chem. Chem. Phys.* **2010**, *12*, 3573–3578.
- (a) Antolínez, S.; Lesarri, A.; Mata, S.; Blanco, S.; López, J. C.; Alonso, J. L. *J. Mol. Struct.* **2002**, *612*, 125–131. (b) Lesarri, A.; Mata, S.; López, J. C.; Alonso, J. L. *Rev. Sci. Instrum.* **2003**, *74*, 4799–4804. (c) Lesarri, A.; Mata, S.; Blanco, S.; López, J. C.; Alonso, J. L. *J. Chem. Phys.* **2004**, *120*, 6191–6196.
- Lesarri, A.; Cocinero, E. J.; López, J. C.; Alonso, J. L. *J. Am. Chem. Soc.* **2005**, *127*, 2572–2579.
- Lesarri, A.; Cocinero, E. J.; López, J. C.; Alonso, J. L. *Chem. Phys.* **2005**, *6*, 1559–1566.
- (a) Brown, R. D.; Godfrey, P. D.; Storey, J. W. V.; Bassez, M. P. *J. Chem. Soc., Chem. Commun.* **1978**, 547–548. (b) Suenram, R. D.; Lovas, F. J. *J. Mol. Spectrosc.* **1978**, *72*, 372–382. (c) Suenram, R. D.; Lovas, F. J. *J. Am. Chem. Soc.* **1980**, *102*, 7180–7184. (d) Lovas, F. J.; Kawashima, Y.; Grabow, J.-U.; Suenram, R. D.; Fraser, G. T.; Hirota, E. *Astrophys. J.* **1995**, *455*, L201–L204. (e) Godfrey, P. D.; Brown, R. D. *J. Am. Chem. Soc.* **1995**, *117*, 2019–2023. (f) McGlone, S. J.; Elmes, P. S.; Brown, R. D.; Godfrey, P. D. *J. Mol. Struct.* **1999**, *485*, 225–238.
- Godfrey, P. D.; Firth, S.; Hatherley, L. D.; Brown, R. D.; Pierlot, A. P. *J. Am. Chem. Soc.* **1993**, *115*, 9687–9691.
- Cocinero, E. J.; Lesarri, A.; Sanz, M. E.; López, J. C.; Alonso, J. L. *Chem. Phys. Chem.* **2006**, *7*, 1481–1487.
- Sanz, M. E.; Cortijo, V.; Caminati, W.; López, J. C.; Alonso, J. L. *Chem.—Eur. J.* **2006**, *12*, 2564–2570.
- Godfrey, P. D.; Brown, R. D.; Rodgers, F. M. *J. Mol. Struct.* **1996**, *376*, 65–81.
- (a) Pirrung, M. C. *Acc. Chem. Res.* **1999**, *32*, 711–718. (b) Salaün, J. *Top. Curr. Chem.* **2000**, *207*, 1–67. (c) Brackmann, F.; de Meijere, A. *Chem. Rev.* **2007**, *107*, 4493–4537.
- (a) Burgess, K.; Ho, K.-K.; Pettitt, B. M. *J. Am. Chem. Soc.* **1995**, *117*, 54–65. (b) Moye-Sherman, D.; Jin, S.; Ham, I.; Lim, D.; Scholtz, J. M.; Burgess, K. *J. Am. Chem. Soc.* **1998**, *120*, 9435–9443. (c) Jiménez, A. I.; Cativiela, C.; Aubry, A.; Marraud, M. *J. Am. Chem. Soc.* **1998**, *120*, 9452–9459. (d) Jiménez, A. I.; Ballano, G.; Cativiela, C. *Angew. Chem., Int. Ed.* **2005**, *44*, 396–399. (e) Royo, S.; De Borggraeve, W. M.; Peggion, C.; Formaggio, F.; Crisma, M.; Jiménez, A. I.; Cativiela, C.; Toniolo, C. *J. Am. Chem. Soc.* **2005**, *127*, 2036–2037. (f) Crisma, M.; Toniolo, C.; Royo, S.; Jiménez, A. I.; Cativiela, C. *Org. Lett.* **2006**, *8*, 6091–6094.
- (a) de Meijere, A. *Angew. Chem., Int. Ed. Engl.* **1979**, *18*, 809–826. (b) Rademacher, P. *Chem. Rev.* **2003**, *103*, 933–975.
- Kohler, E. P.; Conant, J. B. *J. Am. Chem. Soc.* **1917**, *39*, 1404–1420.
- Carr, E. P.; Burt, C. P. *J. Am. Chem. Soc.* **1918**, *40*, 1590–1600.
- (a) Coulson, C. A.; Moffitt, W. E. *Philos. Magn.* **1949**, *40*, 1–35. (b) Walsh, A. D. *Trans. Faraday Soc.* **1949**, *45*, 179–190.
- (a) Pete, J. P. *Bull. Soc. Chim. Fr.* **1967**, 357–370. (b) Cromwell, N. H.; Graff, M. A. *J. Org. Chem.* **1952**, *17*, 414–424. (c) Dauben, W. G.; Berezin, G. H. *J. Am. Chem. Soc.* **1967**, *89*, 3449–3452. (d) Monti, S. A. *J. Org. Chem.* **1970**, *35*, 380–383. (e) Van-Catledge, F. A.; Boerth, D. W.; Kao, J. J. *J. Org. Chem.* **1982**, *47*, 4096–4106.

- (27) Allen, F. H. *Acta Crystallogr. Sect B* **1980**, *36*, 81–96.
- (28) (a) Bartell, L. S.; Guillory, J. P. *J. Chem. Phys.* **1965**, *43*, 647–653. (b) Hoffmann, R. *Tetrahedron Lett.* **1965**, *43*, 3819–3824. (c) Volltrauer, H. N.; Schwendeman, R. H. *J. Chem. Phys.* **1971**, *54*, 260–267. (d) de Mare, G. R.; Peterson, M. R. *J. Mol. Struct.* **1983**, *104*, 115–130. (e) Pawar, D. M.; Noe, E. A. *J. Org. Chem.* **1998**, *63*, 2850–2853. (f) Durig, J. R.; Shen, S. *Spectrochim. Acta, A* **2000**, *56*, 2545–2561. (g) Durig, J. R.; Yu, Z.; Shen, S.; Warren, R.; Verma, V. N.; Guirgis, G. A. *J. Mol. Struct.* **2001**, *563*, 141–145. (h) Hou, X.-J.; Huang, M.-B. *J. Mol. Struct. (Theochem.)* **2002**, *585*, 93–104. (i) Rademacher, P.; Coskum, T.; Kowski, K.; Larionov, O.; de Meijere, A. *Chem.—Eur. J.* **2003**, *9*, 2953–2962. (j) Kazuta, Y.; Abe, H.; Yamamoto, T.; Matsuda, A.; Shuto, S. *Tetrahedron* **2004**, *60*, 6689–6703. (k) Wurrey, C. J.; Zheng, C.; Guirgis, G. A.; Durig, J. R. *Phys. Chem. Chem. Phys.* **2004**, *6*, 2125–2135. (l) Dian, B. C.; Brown, G. G.; Douglass, K. O.; Pate, B. H. *Science* **2008**, *320*, 924–928.
- (29) (a) Powell, D. L.; Klaboe, P. *J. Mol. Struct.* **1973**, *15*, 217–223. (b) Marstokk, K.-M.; Mollendal, H.; Samdal, S. *Acta Chem. Scand.* **1991**, *45*, 37–45. (c) Badawi, H. M.; Al-Saadi, A. A.; Al-Khaldi, M. A. A.; Al-Abbad, S. A.; Al-Sunaidi, Z. H. A. *Spectrochim. Acta, A* **2008**, *71*, 1540–1546.
- (30) (a) Bartell, L. S.; Guillory, J. P.; Parks, A. T. *J. Phys. Chem.* **1965**, *69*, 3043–3048. (b) Durig, J. R.; Bist, H. D.; Little, T. S. *J. Mol. Struct.* **1984**, *116*, 345–359. (c) Wang, A.; Little, T. S.; Durig, J. R. *Spectrochim. Acta, A* **1994**, *50*, 595–607. (d) Durig, J. R.; Shen, S.; Drew, B. R.; Zhao, W. *Struct. Chem.* **2000**, *11*, 213–228. (e) Kazuta, Y.; Abe, H.; Yamamoto, T.; Matsuda, A.; Shuto, S. *J. Org. Chem.* **2003**, *68*, 3511–3521.
- (31) (a) Volltrauer, H. N.; Schwendeman, R. H. *J. Chem. Phys.* **1971**, *54*, 268–273. (b) Noe, E. A.; Young, R. M. *J. Am. Chem. Soc.* **1982**, *104*, 6218–6220. (c) Durig, J. R.; Wang, A.-Y.; Little, T. S. *J. Mol. Struct.* **1991**, *244*, 117–137. (d) Durig, J. R.; Wang, A.-Y.; Little, T. S. *J. Mol. Struct.* **1992**, *269*, 285–308.
- (32) Buñuel, E.; Cativiela, C.; Díaz de Villegas, M. D.; Jiménez, A. I. *Synlett.* **1992**, 579–581.
- (33) Frisch, M. J.; et al. Gaussian 03, revision B.04; Gaussian, Inc.: Pittsburgh, PA, 2003.
- (34) Watson, J. K. G. In *Vibrational Spectra and Structure*; Durig, J. R., Ed.; Elsevier: Amsterdam, 1977; Vol. 6, pp 1–89.
- (35) Gordy, W.; Cook, R. L. *Microwave Molecular Spectra*; Wiley: New York, 1984.
- (36) Pickett, H. M. *J. Mol. Spectrosc.* **1991**, *148*, 371–377.
- (37) (a) Ruoff, R. S.; Klots, T. D.; Emilsson, T.; Gutowsky, H. S. *J. Chem. Phys.* **1990**, *93*, 3142–3150. (b) Miller, T. F.; Clary, D. C.; Meijer, A. J. H. M. *J. Chem. Phys.* **2005**, *122*, 244323.
- (38) Sanz, M. E.; Lesarri, A.; Peña, M. I.; Vaquero, V.; Cortijo, V.; López, J. C.; Alonso, J. L. *J. Am. Chem. Soc.* **2006**, *128*, 3812–3817.
- (39) Blanco, S.; López, J. C.; Mata, S.; Alonso, J. L. *Angew. Chem., Int. Ed.* **2010**, *49*, 9187–9192.
- (40) (a) Császár, A. G. *J. Am. Chem. Soc.* **1992**, *114*, 9568–9575. (b) Miller, T. F.; Clary, D. C. *Phys. Chem. Chem. Phys.* **2004**, *6*, 2563–2571.
- (41) (a) Durmaz, S.; Kollmar, H. *J. Am. Chem. Soc.* **1980**, *102*, 6942–6945. (b) Clark, T.; Spitznagel, G. W.; Klose, R.; Schleyer, P. R. *J. Am. Chem. Soc.* **1984**, *106*, 4412–4419. (c) Rademacher, P. *ChemPhysChem* **2003**, *4*, 1237–1240.
- (42) (a) Hendricksen, D. K.; Harmony, M. D. *J. Chem. Phys.* **1969**, *51*, 700–705. (b) Kalasinsky, V. F.; Powers, D. E.; Harris, W. C. *J. Phys. Chem.* **1979**, *83*, 506–510. (c) Compton, D. A. C.; Rizzolo, J. J.; Durig, J. R. *J. Phys. Chem.* **1982**, *86*, 3746–3753. (d) Rall, M.; Harmony, M. D.; Cassada, D. A.; Staley, S. W. *J. Am. Chem. Soc.* **1986**, *108*, 6184–6189. (e) Diallo, A. O.; Van-Thanh, N.; Rossi, I. *Spectrochim. Acta, A* **1987**, *43*, 415–420. (f) Iijima, T.; Kondou, T.; Takenaka, T. *J. Mol. Struct.* **1998**, *445*, 23–28. (g) Hyla-Kryspin, I.; Grimme, S.; Hruschka, S.; Haufe, G. *Org. Biomol. Chem.* **2008**, *6*, 4167–4175.
- (43) Császár, A. G.; Allen, W. D.; Schaefer, H. F. *J. Chem. Phys.* **1998**, *108*, 9751–9764.