J. Am. Chem. Soc. 1984, 106, 6243-6245

Quantum and Molecular Mechanical Studies on Alanyl Dipeptide

Scott J. Weiner, U. Chandra Singh, T. J. O'Donnell,† and Peter A. Kollman*

Contribution from the Department of Pharmaceutical Chemistry, University of California, San Francisco, California 94143, and Abbott Laboratories, North Chicago, Illinois 60064.
Received January 3, 1984

Abstract: Molecular mechanical and ab initio (4-31G) calculations on N^{α}-acetyl-N-methylalaninamide have been carried out. At the molecular mechanical level, five local energy minima have been found, and the free energies have also been determined for these five structures. Addition of a dispersion energy term to the SCF quantum mechanical energies has been shown to affect the relative energies of these local minima and to improve the agreement between the quantum mechanical and molecular mechanical relative energies. The structural properties of the five conformers calculated at the molecular mechanical and quantum mechanical level are in generally good agreement.

Many theoretical[1-7] and experimental[8-11] studies have been performed on the potential surface of alanyl dipeptide. Most recently, Scarsdale et al.[3] have performed ab initio quantum mechanical (4-21G basis set) geometry optimization on seven N^{α}-acetyl-N-methylalaninamide (NANA) structures and have compared their results to ECEPP molecular mechanical calculations. In our opinion, their work has given a somewhat incomplete view of the "state of the art" of molecular mechanical calculations on this molecule. Thus, the goals of this paper are threefold: first, to suggest that "state of the art" molecular mechanical and quantum mechanical calculations are in *qualitative* agreement with each other, provided that one relaxes all geometrical degrees of freedom and considers dispersion attraction; second, to evaluate basis set differences at the ab initio level on the relative energies of some of the conformations of NANA; and, finally, to present relative free energies of the local minima on the NANA surface.

Methods

To explore the NANA potential energy surface, we used our molecular mechanics program AMBER[12] and the technique of adiabatic mapping, where the Φ, Ψ dihedrals were constrained to fixed values and the remaining degrees of freedom allowed to relax. We have only considered trans peptide units in this study. The potential energy function, parameter set, and Φ, Ψ plot appear in ref 6. All low-energy structures were subjected to further refinement by using a second-derivative Newton-Raphson algorithm to assess whether they were local minima. The alanyl dipeptide unit appears in Figure 1. Two local minima conformations correspond to 1-7 hydrogen-bonded systems between O6 and H18. These are $\Phi \approx -75°$, $\Psi \approx 65°$ (C_7^{eq}) and $\Phi \approx 70°$, $\Psi \approx -65°$ (C_7^{ax}). A third local minima conformation, with a 1-5 hydrogen bond between O16 and H8, (C_5) has $\Phi \approx -160°$, $\Psi \approx 170°$. Two other local minima found are characterized by $\Phi \approx -60°$, $\Psi \approx -40°$ (C_{10}^{R}) and $\Phi \approx 55°$, $\Psi \approx 45°$ (C_{10}^{L}). The geometries of the three lowest energetic local minima (C_7^{eq}, C_7^{ax}, and C_5) were taken as starting structures for quantum mechanical ab initio (4-31G basis set) gradient optimization by using the program GAUSSIAN 80 U.C.S.F.[13] To evaluate the dispersion correction (DC) term for the quantum mechanical results, a $-B/R^6$ term was used (B values appear in ref 6). We have reduced the 1-4 dispersion interactions by a factor of 2, consistent with our molecular mechanical model; this reduction of short distance dispersion attraction also has precedence in ref 14. All calculations were performed on VAX 11-780's, with each quantum mechanical calculation taking ≈20 h for SCF + gradient evaluation.

Results and Discussion

The results of our molecular mechanical and quantum mechanical calculations appear in Tables I–III. Since ab initio SCF calculations do not contain dispersion energy,[15] which could lead to differences in the conformational energies, we have amended the Scarsdale et al.[3] results with a dispersion-energy correction (DC) (Table I). The DC for the 4-21G alanyl structures is −10.8, −11.5, −9.7, −9.6, and −11.2 kcal/mol for C_7^{eq}, C_7^{ax}, C_5, C_{10}^{R}, and C_{10}^{L}, respectively (summing over all nonbonded interactions pairs).

The addition of these DC values to the 4-21G ab initio energies significantly improves the agreement between the quantum mechanical and molecular mechanical relative energies. First, the ordering of the C_7^{ax} and C_5 structures, relative to C_7^{eq}, is reversed (Table I). Stern et al.[5] and our gas-phase molecular mechanical models ($\epsilon = 1$) find C_7^{eq} to be more stable than C_7^{ax} by 1.1–1.3 kcal/mol, in reasonable agreement with (4-21G) QM + DC of 1.7 kcal/mol. The $C_7^{eq} - C_5$ energy difference found in our molecular mechanical model is not as close to the QM + DC value (4.0 vs. 2.5 kcal/mol).[16] Finally, the other two local minima found here are reasonably close in energy to the corresponding ab initio ones, although for C_{10}^{R}, the ab initio structure differs somewhat from the molecular mechanical one, likely due to the shallowness of the potential surface in this region.[3]

It is clear that the dispersion correction is an important addition to the ab initio relative energies. Other evidence for the importance of DC in evaluating relative energies comes from a study of the corresponding glycyl dipeptide C_7 and C_5 structures at the ab initio 4-21G level, where an energy difference of 0.8 kcal/mol was found. For the glycyl peptide the DC is −8.5 kcal/mol for C_7 and −6.7 kcal/mol for C_5. When the DC is added to these structures, the energy difference becomes 2.5 kcal/mol, in closer agreement with the molecular mechanics ($\epsilon = 1$) difference of 3.9 kcal/mol. While we cannot claim that our dispersion correction is quantitatively

---

* University of California.

† Abbott Laboratories.

(1) Pullman, B.; Pullman, A. *Adv. Protein Chem.* **1974**, *28*, 348.

(2) Zimmerman, S.; Pottle, M.; Nemethy, G.; Scheraga, H. *Macromolecules* **1977**, *10*, 1.

(3) Scarsdale, J.; Van Alsenoy, C.; Klimkowski, V.; Schafer, L.; Momany, F. *J. Am. Chem. Soc.* **1983**, *105*. 3438.

(4) Brooks, B.; Bruccoleri, R.; Olafson, B.; States, D.; Swaminathan, S.; Karplus, M. *J. Compt. Chem.* **1983**, *4* 187.

(5) Stern, P.; Chorev, M.; Goodman, M.; Hagler, A. *Biopolymers* **1983**, *22*, 1885.

(6) Weiner, S.; Kollman, P.; Singh, U.; Case, D.; Ghio, C.; Alagona, G.; Profeta, S.; Weiner, P. *J. Am. Chem. Soc.* **1984**, *106*, 765.

(7) Peters, D.; Peters, J. *J. Mol. Struct.* **1981**, *85*, 107.

(8) Cung, M.; Marraud, M.; Neel, J. In "The Jerusalem Symposia on Quantum Chemistry and Biochemistry—Conformation of Biological Molecules and Polymers"; Bergmann, E., Pullman, B., Eds.; Reidel: Boston, 1973; p 69.

(9) Avignon, M.; Lascombe, J., ref 8, p 97.

(10) Avignon, M.; Van Huong, P. *Biopolymers* **1970**, *9*, 427.

(11) Burgess, A.; Scheraga, H. *Biopolymers* **1973**, *12*, 2177.

(12) Weiner, P.; Kollman, P. *J. Compt. Chem.* **1981**, *2*, 287.

(13) Singh, U. C.; Kollman, P. *QCPE* **1982**, *2*, 17 (No. 446).

(14) See Douketis et al. (Douketis, C.; Scoles, G.; Marchetti, S.; Zen, M.; Thakkar, A. *J. Chem. Phys.* **1982**, *76*, 3057) for a leading reference to calculate dispersion energy as a series of R^{-6}, R^{-8}, and R^{-10} terms.

(15) Margenau, H.; Kestner, N. "The Theory of Intermolecular Forces"; Pergamon Press: Oxford, 1970.

(16) There is no local minimum in the Hagler et al. surface for the C_5 structure, although such a structure appears to be ≈3–4 kcal/mol higher in energy (estimated from their contour plot).

0002-7863/84/1506-6243$01.50/0

Table I. Structures and Energies for Alanyl Dipeptide

	FF2[a] $\epsilon = 1$	FF2[a] $\epsilon = 4$	FF2[a] $\epsilon = R_{i,j}$	4-31G[b] QM+DC	4-21G[c] QM+DC	ECEPP[d] $\epsilon = 2$	ref 5[e] $\epsilon = 1$	PCILO[f]
ϕ^g	−75.6	−80.4	−75.6	−81.1	−84.6	−84	−80	−78
Ψ^g	68.5	69.0	65.6	66.3	73.0	79	80	40
ΔE^h	0.0	0.0	0.0	0.0 (0.0)	0.0 (0.0)	0.0	0.0	0.3
ΔG^h	0.0	0.0	0.0					
Φ	68.6	72.1	69.0	73.6	74.6	78	70	75
Ψ	−67.4	−68.2	−64.4	−60.4	−62.0	−64	−80	−40
ΔE	1.1	0.7	0.6	2.2 (2.4)	1.7 (2.6)	8.8	1.3	0.0
ΔG	1.2	0.9	0.7					
Φ	−157.6	−159.3	−160.7	−161.8	−165.7	−154		−171
Ψ	169.1	166.1	168.6	167.3	167.3	153		164
ΔE	4.0	1.2	3.2	1.9 (0.2)	2.5 (1.4)	0.4		1.6
ΔG	2.9	0.3	2.1					
Φ	−60.3	−65.2	−60.7		−91.9	−74	−60	−29
Ψ	−34.2	−50.1	−40.7		−5.5	−45	−40	−59
ΔE	4.7	1.2	3.6		6.1 (4.9)	1.1	4.2	2.4
ΔG	3.6	0.3	2.6					
Φ	53.5	55.8	53.9		60.8	54	60	
Ψ	36.5	50.8	41.6		40.6	57	40	
ΔE	5.7	1.8	4.3		6.3 (6.7)	2.3	7.3	
ΔG	4.9	1.3	3.6					

[a] Molecular mechanical simulations using the program AMBER (ref 12) and second-derivative techniques to ensure that each of the five structures was a "true" local minimum. FF2 parameters appear in ref 6. The dielectric constant used appears in each column as ϵ. [b] This study, gradient optimized using GAUSSIAN 80 U.C.S.F. (ref 13). The total energy for C_7^{eq} is −492.133696 au. The largest gradient component for the three structures C_7^{eq}, C_7^{ax}, and C_5 are 0.0015, 0.0068, and 0.0013 au, respectively. Over the last two optimization cycles the energy was reduced 0.04, 0.04, and 0.003 kcal/mol for C_7^{eq}, C_7^{ax}, and C5 respectively. The ΔE = QM + DC in kcal/mol. The values in parentheses correspond to the QM values. [c] Structures are from Scarsdale et al. (ref 3). [d] Empirical energy calculations by Zimmerman et al. using fixed bond lengths and angles (ref 2). [e] molecular mechanics refined structures by Stern et al. Each of the Φ, Ψ values was constrained in 10° increments with all other degrees of freedom allowed to relax (ref 5). [f] PCILO results by Pullman et al. (ref 1). [g] The specific dihedral angles are illustrated in Figure 1. For the proper convention see ref 6. [h] All energies ΔG and ΔE are in kcal/mol.

Table II. Bond Angles around C_α in NANA (deg)

structure	molecular mechanics (ϵ = 1)[a]					
	$N-C_\alpha-C_\beta$	$N-C_\alpha-C$	$N-C_\alpha-H_\alpha$	$C_\beta-C_\alpha-C$	$C_\beta-C_\alpha-H_\alpha$	$H_\alpha-C_\alpha-C$
C_7^{eq}	110.5	109.8	109.0	112.6	107.0	107.9
C_7^{ax}	112.9	111.1	106.2	113.2	106.2	106.7
C_5	111.1	107.7	108.9	111.9	108.8	108.3
C_{10}^R	110.7	111.4	108.1	111.3	107.7	107.5
C_{10}^L	111.1	111.8	107.1	112.8	107.0	106.6
			4-21G[b]			
C_7^{eq}	110.1	109.5	106.8	110.6	111.1	108.6
C_7^{ax}	112.4	112.7	106.0	111.8	109.1	104.2
C_5	111.6	106.4	109.3	110.5	109.3	109.8
C_{10}^R	110.6	114.0	108.0	108.4	109.8	106.0
C_{10}^L	113.3	111.1	105.8	110.6	109.7	106.0
			4-31G[a]			
C_7^{eq}	109.7	110.5	107.2	111.3	110.1	108.0
C_7^{ax}	113.3	112.8	105.3	111.9	108.6	104.3
C_5	111.8	107.2	108.8	110.9	108.8	109.3

[a] This study. [b] Reference 3.

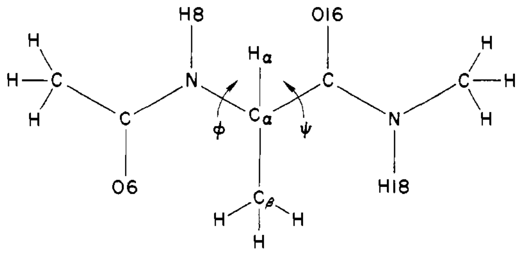

Figure 1.

accurate,[14] it is likely that the C_7^{ax} and C_7^{eq} structures will be stabilized relative to C_5 no matter what the dispersion coefficients. Further support for the reasonableness of the approach taken here is found in the work of Prissette and Kochanski.[17] They have shown that for different configurations of $(Cl_2)_2$, a simple atom centered $-B/R^6$ dispersion term does an excellent job of reproducing a more complete quantum mechanical calculation of dispersion attraction between the two chlorine molecules.

Scarsdale et al.[3] pointed out that the molecular mechanical (ECEPP) calculations of Zimmerman et al.[2] were inconsistent with their quantum mechanical calculations, because ECEPP finds C_7^{ax} to be 8.8 kcal/mol higher in energy than C_7^{eq}, compared with only 1.7 kcal/mol for the (4-21G) QM + DC results. This difference is caused by ECEPP's use of fixed bond lengths and angles, since molecular mechanical results which allow full relaxation of these parameters are consistent with the quantum mechanical values.[4-6] The rigid geometry ab initio STO-3G energies of Peters and Peters[7] also differ significantly from the relaxed geometry ab initio and molecular mechanical values.

We had begun a 4-31G gradient optimization on the C_7^{eq}, C_7^{ax}, and C_5 structures of NANA, beginning with the molecular mechanics refined geometries, before the Scarsdale et al.[3] work

---

(17) Prissette, J.; Kochanski, E. *J. Am. Chem. Soc.* **1978**, *100*, 6609.

Table III. Representative Dihedral Angles for NANA (deg)

structure	CH1–C–N–C_α	C–N–C_α–C	N–C_α–C–N	C_α–C–N–CH2
		Molecular Mechanics (ϵ = 1)[a]		
C_7^{eq}	178.4	−75.5	68.5	−179.9
C_7^{ax}	−177.8	68.6	−67.5	−179.8
C_5	177.0	−157.6	169.0	179.6
C_{10}^R	−179.2	−60.3	−34.4	179.5
C_{10}^L	178.8	53.7	36.6	−178.9
		4-21G[b]		
C_7^{eq}	−177.0	−84.6	73.0	−179.0
C_7^{ax}	176.0	74.7	−62.0	−178.9
C_5	178.7	−165.7	167.3	178.0
C_{10}^R	−173.1	−91.9	−5.5	179.0
C_{10}^L	174.5	60.8	40.6	178.4
		4-31G[a]		
C_7^{eq}	−178.6	−81.1	66.3	−178.7
C_7^{ax}	178.2	73.6	−60.4	−179.8
C_5	178.7	−161.8	167.3	178.8

[a] This study. [b] Reference 3.

appeared (Table I). The relative energies of C_7^{eq} and C_5 before dispersion correction are surprisingly different (0.2 kcal/mol for 4-31G, 1.4 kcal/mol for 4-21G), whereas the $C_7^{eq} - C_7^{ax}$ differences are nearly identical (2.4 kcal/mol for 4-31G, 2.6 kcal/mol for 4-21G). We confirmed that this $C_7^{eq} - C_5$ difference was *not* mainly due to geometric differences by carrying out 4-31G single-point ab initio calculations on C_5 and C_7^{eq} at the 4-21G geometries, finding $\Delta E_{C_7^{eq}-C_5} = 0.6$ kcal/mol. Thus, the remaining energy difference of 0.8 kcal/mol between such similar basis sets remains a mystery.[18] The 4-31G ab initio energy difference between C_5 and C_7^{ax} is large enough that even dispersion correction does not reverse the order of stabilities, even though it brings them closer.

We have compared the bond lengths, bond angles, and dihedral angles found in the molecular mechanical and 4-31G and 4-21G quantum mechanical calculations on the various conformations of NANA. The calculated bond lengths are insensitive to conformation and are very similar in the three sets of calculations, with standard deviations among the calculated values for given bonds ranging from 0.003 to 0.01 Å. For the bond angles (looking now only at the three conformations C_7^{ax}, C_7^{eq}, and C_5 for each angle; this corresponds to 36 angles for each conformation for a total of 108 total bond angles) 77 are found with standard deviations less than 1.0°, 30 with standard deviations between 1.0° and 2.0°, and 1 with a standard deviation of 2.5°. The largest conformation dependent angle differences are around the C_α carbon, and, thus, we report these in detail in Table II.

Focusing on the C_α angles for the three lowest energy structures (Table II), we see that the agreement between the quantum mechanical and molecular mechanical calculated values is good for those angles not involving hydrogens (N–C_α–C_β, N–C_α–C, and C_β–C_α–C) with the order of the angles identical for both calculations and the values of the angles differing on average by 1.0° (largest discrepancy being 2°). There are larger differences between the calculated angles involving hydrogens (N–C_α–H_α, C_β–C_α–H_α, and C–C_α–H_α), with an average difference of 1.7° and the largest difference 4.1°. Given that the force field reported in ref 6 has been optimized for united atoms, and not as yet at the all-atom level, the agreement is still reasonable. It is likely that the quantum mechanical calculations are more quantitatively accurate in the calculations of these angles, but it is not clear that the extra accuracy is worth the computational price here, given the 3–4 order of magnitude difference in computer time involved in the two types of calculations.

The Φ, Ψ and peptide (ω) dihedral angles are compared in Table III. The Φ, Ψ values are qualitatively similar for a given conformation with the exception of C_{10}^R. As Scarsdale et al.[3] point out, however, the potential surface as a function of these dihedral angles is very shallow and the 4-21G and 4-31G structures are not necessarily at true local minima. The two ω dihedral angles are similar and differ by at most 6° between 4-21G and molecular mechanics, but the two methods do not always agree on the sign of the deviation from planarity.

A final factor which must be considered in relating the calculated results to experimental conformer populations are entropy differences. We can approximate the relative gas-phase entropies and free energies for the molecular mechanics models within the rigid rotor, harmonic oscillator approximations for various conformations.[20] The entropic contribution at 298 K helps stabilize the C_5 conformation, relative to either C_7, by ≈1 kcal/mol (Table I).

Discussion

It is clear that there is no major qualitative discrepancy between the dispersion corrected ab initio results and the molecular mechanical for gas-phase NANA. The gas-phase (ϵ = 1) molecular mechanical results reported by us and Stern et al.[5] and the 4-21G (and 4-31G) QM + DC find C_7^{eq} lowest in energy, C_7^{ax} higher by 1–2 kcal/mol, C_5 higher by 2–4 kcal/mol, and C_{10}^R, C_{10}^L higher by 5–7 kcal/mol. The largest quantitative discrepancy between the relative molecular mechanical and quantum mechanical energies is the magnitude of the energy difference between C_7^{eq} and C_5 which is 1.9–2.5 kcal/mol at the ab initio level and 3–4 kcal/mol at the molecular mechanical level. The quantum mechanical calculations still suffer from basis set dependence and too simple dispersion correction, so they are not yet a definitive reference point. Experimental data on NANA in nonpolar solvents are most consistent with a nonnegligible fraction of C_5, suggesting that the quantum mechanical values are closer to correct. On the other hand, we have demonstrated that both entropy effects and dielectric constant changes profoundly affect the magnitude of the $C_7^{eq} - C_5$ energy difference at the molecular mechanical level, with the C_7^{eq} and C_5 of approximately equal free energies with ϵ = 4. This, combined with the uncertainty in the experimental data in nonpolar solvents, precludes a definitive evaluation of how far from the truth these various calculated energy differences are. Thus, it is important to perform further gas-phase experiments on this most important molecule to definitively establish its conformational equilibrium.

Acknowledgment. P.A.K. thanks the NIH (GM-29072) for research support.

Registry No. NANA, 19701-83-8.

---

(18) Single-point ab initiio calculations on the Schafer et al. (ref 19) glycyl dipeptide structures also give a 0.8-kcal/mol stabilization of C_5 relative to C_7 (4-31G basis set compared to the 4-21G results), such that at the 4-31G level with no dispersion correction, both C_7 and C_5 are approximately equo-energetic.

(19) Schafer, L.; Alsenoy, C.; Scarsdale, J. *J. Chem. Phys.* **1982**, *76*, 1439.

(20) Hagler, A.; Stern, P.; Sharon, R.; Becker, R.; Becker, J.; Naider, F. *J. Am. Chem. Soc.* **1979**, *101*, 6842.