Marked Increments of Stability and Proton Affinity of the Protonated, Zwitterionic Glycine Induced by the Attachment of Two Excess Electrons

Hongqi Ai,[†] Yuxiang Bu,*[,†,‡] Ping Li,[†] and Lixiang Sun[†]

Institute of Theoretical Chemistry, Shandong University, Jinan, 250100, P. R. China, and Department of Chemistry, Qufu Normal University, Qufu, 273165, P. R. China

Received: February 9, 2004; In Final Form: March 8, 2004

The most stable protonated glycine (^{1}GlyH1) attached by different charges (0, ±1, ±2, −3, and −4) in the gas phase has been comprehensively studied by using density functional theory (DFT/B3LYP) and the CCSD method. Results show that, on the basis of protonation, the zwitterionic glycine (GlyZW) can be further stabilized with a 90.4 kcal/mol energy drop by attaching an excess electron in the dipole-bound mode (^{2}GlyH0). The corresponding vertical electron affinity is −86.9 kcal/mol. Interestingly, two-electron-attached ^{1}GlyH1 [^{1}GlyH(−1)] is more stable by 5.4 kcal/mol than the one-electron-attached one (^{2}GlyH0). The analyses for the probable dissociation modes of the series ^{m}GlyHn (n, charge; m, spin multiplicity) species also confirm this conclusion. The additional stability mainly stems from the contribution of the deformation energy induced by the two excess electrons. Results show that the deformation contribution induced by attaching one, two, or three electrons can favor the stability of each corresponding system, while either attaching four electrons or ionizing one electron would make the corresponding system unstable. On the other hand, the greater the number of attached electrons is, the more the GlyZW species combines a proton readily, i.e., with larger proton affinity (PA). For example, the CCSD(T)-calibrated PA of ^{2}GlyZW(−1) is 316.9 kcal/mol, larger by 104.3 kcal/mol than that of ^{1}Gly0. The PA of ^{1}Gly0 calibrated at the same level in the present paper is 212.6 kcal/mol, in excellent agreement with the theoretical (211.1 kcal/mol) and experimental (211.8 kcal/mol) results.

Introduction

It is known that the amino acids and peptides form the building blocks of proteins. Among biomolecules, peptides are unique in having seemingly infinite conformational possibilities and a broad spectrum of functional groups. Three-dimensional structural information on simpler species such as the isolated amino acids[1,2] is very significant for one to gain a good understanding of the biological activity of a peptide.

While amino acids in solution exist as zwitterions predominately at pH 7, with the carboxyl deprotonated and one of the nitrogen atoms protonated, the most stable form of isolated amino acids in the gas phase is nonzwitterionic. As the simplest amino acid, glycine has been extensively studied by theoretical and experimental methods.[3−18] Its nonzwitterionic form is calculated to be 19.3 kcal/mol more stable than the zwitterionic one (GlyZW)[3]. Both millimeter wave spectroscopy experiments[19] and the gas-phase basicity measurements[20] have demonstrated that glycine is not a zwitterion in the gas phase. Because the information about the gas-phase intrinsic properties can play a large role in solution-phase measurements, a number of efforts have been made in stabilizing and characterizing the gas-phase zwitterionic ions. These efforts have concentrated on hydrated,[21−23] metal ion cationized,[24−28] protonated,[24,29−35] and one-electron-attached glycine derivatives.[36] For the hydrated GlyZW, the dipole interaction between the glycine and the water molecules is the main contribution to the stability of their complex due to the large dipole moment of GlyZW,[37] though there is not net charge on itself. Jensen et al.[38] have determined that two water molecules can stabilize the glycine zwitterions. For the metal ion stabilized GlyZW systems, two cases have been observed. One is the monovalence metal ion coupled system, in which the cationization can stabilize GlyZW and produce a local minimum, but not a global one, on its potential energy surface (PES).[24,25,27] Another is the divalent metal ion cationized system, in which the cationization not only stabilizes GlyZW but also yields a complex with a global minimum on its PES.[26−28] In these interactions, the electrostatic effect between the glycine and the metal ion plays a significant role, especially for the interactions in those divalent metal ion coupled systems.[26−28]

Protonation of amino acids has received much interest not only because it can stabilize the zwitterionic amino acids[24,29−35] but also because it is the dominant ionization pathway during the analysis of peptides by mass spectrometry, which is one of the major experimental means to study biomolecules.[39] So for the simplest but representative glycine, studies on its protonation have been extensively performed by many work groups.[24,29−35,40] Both theoretical and experimental results have confirmed that the protonated glycine ^{1}GlyH1 is the global minimum on its PES.[4,34,40] All of these researchers have regarded the protonated glycine as a simple but significant model to study the properties of biological molecules. Another means is to attach an excess electron to stabilize GlyZW.[36] By this means, other biomolecules such as arginine,[41] alanine, proline,[42] and nucleic acid bases[43] have also been confirmed theoretically and experimentally to be stabilized by their strong dipole-bound interactions. Moreover, some small inorganic molecules, such as the water molecule cluster, can be also stabilized by the attachment of an

[†] Shandong University.
[‡] Qufu Normal University.

10.1021/jp049405m CCC: $27.50
Published on Web 04/13/2004

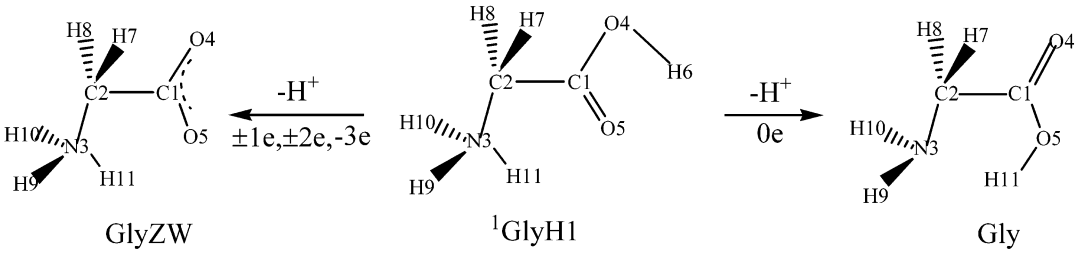

Figure 1. Two dissociation modes of proton for the most stable protonated glycine derivative ^{1}GlyH1 with various charges.

TABLE 1: Relative Energy (adiabatic EA_a, vertical EA_v) of ^{m}GlyH*n* and PA of ^{m}GlyZW(*n*−1)[a]

n, *m*[b]	2, 2		1, 1		0, 2		−1, 1		−2, 2		−3, 1	
DM/method 1	4.3	3.9	5.8		1.5	4.5	7.2	5.9	2.0	2.5	6.3	6.3
ΔZPVE/method 1	−1.6		0.0		−4.0		−5.3		−4.6		−4.7	
PA/method 3	68.5		212.6		316.9		392.4		474.3		−	

	EA_a	EA_v	EA_a	EA_v	EA_a	EA_v	EA_a	EA_v	EA_a	EA_v	EA_a	EA_v
method1	362.0	376.4	0.0	0.0	−99.1	−92.1	−106.2	−93.6	−34.0	−29.8	104.3	107.8
method2	363.0	377.0	0.0	0.0	−99.2	−92.2	−106.2	−93.8	−34.1	−30.4	103.1	106.2
method3	384.5	393.5	0.0	0.0	−90.4	−86.9	−95.8	−88.8	−32.8	−23.3	93.1	95.1

[a] Energy in kcal/mol and dipole moment (DM) in debye. Methods 1, 2, and 3 denote B3LYP/6-31++G**, B3LYP/6-311++G**//B3LYP/6-31++G**, and CCSD(T)/aug-cc-pvdz//B3LYP/6-31++G**, respectively. [b] *n*, *m* denote the charge and corresponding spin multiplicity, respectively.

excess electron.[44] Thus either protonation or an electron attachment can stabilize GlyZW; however, no reports on the combination of the two means to stabilize GlyZW are given, to our best knowledge.

The purpose of this research is to carry out a detailed discussion on how the different charge effects influence the stability of the most stable protonated glycine complex in the gas phase. By the study, we expect to offer an optimal electron number that can efficiently stabilize GlyZW and to show that the combination of both attachment of excess electrons and protonation would be more favorable to the stability of GlyZW than either of the two methods. Moreover, the charge effect on the proton affinity (PA) will also be investigated by analyzing the different electron distributions on each atom of the protonated glycine system. The analyses on the possible dissociation modes of these species are also performed.

Computational Details

Császár[7] had reported 13 stable glycine isomers at higher levels and with larger basis sets previously. In view of the abundance of glycine zwitterionic isomer in the life system and its transformation probability from the neutral structure in the aqueous phase via an intramolecular proton transfer,[26,45,46] the neutral glycine (Gly) and the assumed zwitterionic form GlyZW accompanying ^{1}GlyH1 (see Figure 1) are employed as the starting structure for the discussions in this paper. Note that the suffix *n* and the superscript *m* in ^{m}GlyH*n* denote the charge and corresponding spin multiplicity, respectively.

All these charged GlyH species (^{m}GlyH*n*, $n = 0, \pm1, \pm2$ and -3) are optimized first at the B3LYP/6-31++G** level[47] without any symmetry constraints. For the dipole-bound anion systems, the basis set level has been employed successfully to treat several similar systems.[41,44] Then frequency calculations at the same level are also performed to characterize all of these optimized structures to be genuine minima (i.e., no imaginary frequencies) and to get the zero point vibrational energy (ZPVE) of the corresponding system. Single point calculations with the 6-311++G**[48] basis set are performed to refine these calculated results with the same level. The coupled cluster level[49] with single and double excitations and a perturbative estimate of the triple excitations CCSD(T)[50] and a large basis set (aug-cc-pvdz)[51] are last employed to carry out single point calculation, using the B3LYP optimized geometries, so that these energies are calibrated. ZPVE corrections are included in those PA calculations to compare with the available experimental result.

The DFT methods, in particular the B3LYP one, for the study of the conformational behavior of glycine and other amino acids provide very similar structural parameters as compared with MP2.[52] Moreover, the DFT vibrational frequencies and intensities are in excellent agreement with the experimental data.[45b] So the B3LYP method is first employed to optimize the geometries of these different ^{m}GlyH*n* species and the corresponding charged GlyZW species. As demonstrated by Nguyen et al.,[53] the calculated relative energies of different neutral glycine isomers are sensitive to the level of theory. Besides, electron correlation effects have been proved to be important for dipole-bound anions.[36,41,44,54] Thus CCSD(T) is also employed.

All these calculations are performed with the Gaussian 94 and Gaussian 98 program suites[55] on Origin 300 and Pentium IV/2.4 GHz computers.

Results and Discussions

Because the charge transfer or excursion in a biological system is not limited completely in the one-electron form but also may be in the two-[56−58] or even four-electron[57,59,60] form, the ^{m}GlyH*n* species with $n = 0, \pm1, \pm2$ and -3 were, respectively, designed and optimized. The proton affinity (PAs) of ^{1}Gly0 and these different charged ^{m}GlyZW(*n*−1) ($n \neq 1$) species and calculated relative energies (adiabatic electron affinity energies (EA_as)), relative zero point vibrational energies (ΔZPVE), vertical electron affinity energies (EA_vs), and optimized dipole moments (DMs) of the ^{m}GlyH*n* are listed in Table 1. The selected charges of the ^{m}GlyH*n* species are drawn in Figure 2 and optimized geometries are drawn in Figure 3. No geometries of both the charged Gly and GlyZW are displayed in Figure 3, though they are also optimized with the same level. Only their energies are utilized in the following PA calculations.

Proton affinity is the energy released when a proton is attached to an atom or a molecule. Therefore, the theoretical

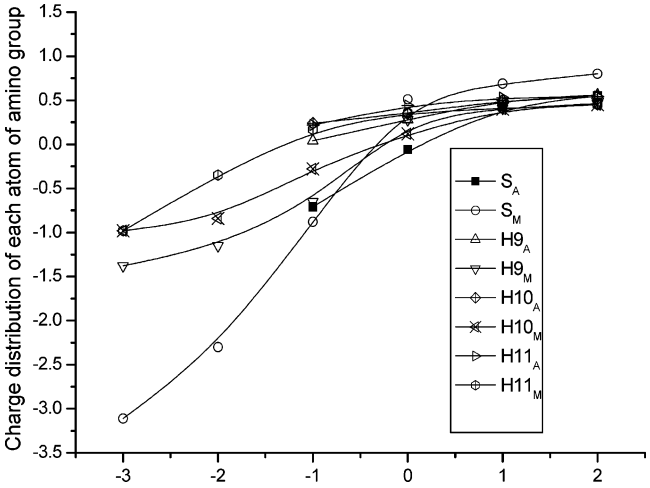

Figure 2. Plots of two charge distributions (S, charge sum of amino group atoms; subscript A, Aim; subscript M, Mulliken) of each atom of the amino group in charged ^{m}GlyH*n*.

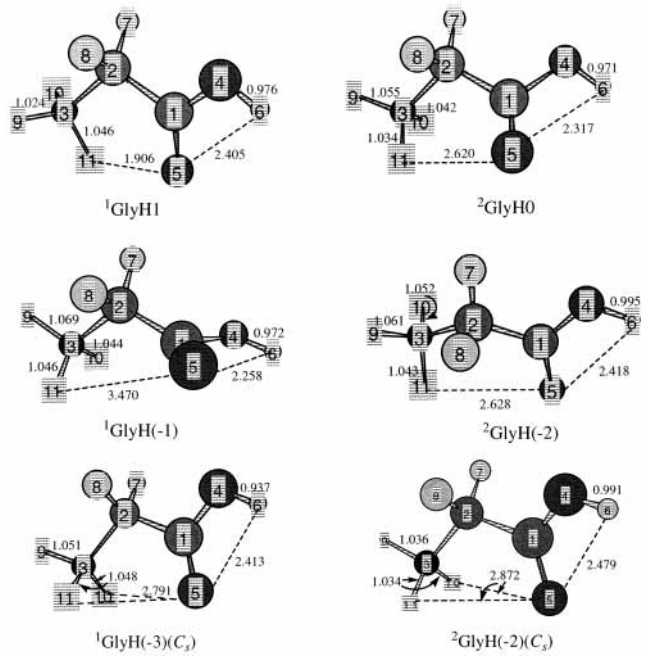

Figure 3. Various B3LYP/6-31++G**-optimized charged ^{m}GlyH*n* derivatives of the most stable protonated glycine ^{1}GlyH1. Distance in angstrom.

PA of ^{m}GlyZW($n-1$) and ^{1}Gly0 in the gas phase is given by[61]

$$\mathrm{PA}[^{m}\mathrm{GlyZW}(n-1)] = -\{E(^{m}\mathrm{GlyH}n) - E[^{m}\mathrm{GlyZW}(n-1)]\} - \Delta\mathrm{ZPVE}' + {}^{5}/_{2}RT \quad (1)$$

Note that ^{m}GlyZW($n-1$) will degenerate into ^{1}Gly0 when $n = 1$, and the ΔZPVE′ is also different from those in the Table 1, in which the ΔZPVE values refer to the differences between those various ^{m}GlyH*n* and ^{1}GlyH1.

Electron affinity (EA) is the energy released when an electron is attached to an atom or a molecule. Therefore, the EA of ^{m}GlyH*n* is given by

$$\mathrm{EA}(^{m}\mathrm{GlyH}n) = -\{E[^{m'}\mathrm{GlyH}(n-1)] - E(^{m}\mathrm{GlyH}n)\} \quad (m \neq m') \quad (2)$$

The corresponding Mulliken charge distributions of each atom are gathered in Table S1 (Supporting Information).[62] To validate the reliability of the Mulliken charge distributions of the ^{m}GlyH*n* species, the theory of atoms in molecules (AIM)[63] and extensions thereto[64] is also employed to recalculate the charge distributions of these atoms on the basis of B3LYP/6-31++G**-optimized structures with the same level. So all the atom charges except those in the ^{2}GlyH(−2) and ^{1}GlyH(−3) species are reobtained and also listed in the Table S1 (see the values in parentheses). Table S1 confirms that the charge changes of amino group hydrogens (including an extra proton) are the major acceptor of excess electrons, and these charge changes are most employed in the following discussion, so they are redrawn in Figure 2 especially to compare them visually. The S in the figure denotes the sum of charges of amino group atoms (N3, H9, H10, and H11). The subscript A and M stand for the results of AIM and Mulliken, respectively. Comparisons between AIM and Mulliken charges of the S or any of three hydrogens show that the tendencies of charge distribution in these different ^{m}GlyH*n* species are consistent. Due to the limitation of AIM, neither the charges of ^{2}GlyH(−2) nor ^{1}GlyH(−3) species can be obtained, so the following discussion will employ only the results from the Mulliken charges.

For these different ^{m}GlyH*n* species, the relative energies obtained at various levels and basis sets, i.e., B3LYP/6-31++G**, B3LYP/6-311++G**, and CCSD(T)/aug-cc-pvdz, are consistent with each other. Especially, a pair of values obtained with the two former methods are almost unchanged however underestimated relative to that obtained at the CCSD-(T) level. As a more reliable value, those obtained at the CCSD-(T) level will be mainly employed in the following discussions, except as specified. It is noted that the ΔZPVE results (take the value of the ^{1}GlyH1 species as the benchmark) of those excess electron-attached ^{m}GlyH*n* species are in the range of −4.0 to −5.3 kcal/mol, which is lower than that (−1.6 kcal/mol) of the further ionized ^{1}GlyH1 (^{2}GlyH2) species (see Table 1). These values can be used as the correction of relative stability of these different ^{m}GlyH*n* systems

Relative Stability and Geometric Characters. *(1) ^{1}GlyH1 and Its Corresponding Two Glycine Isomers.* If we superimpose the Gly and ^{1}GlyH1 molecules and use the same coordinate system, then the two calculated dipoles are 5.9 and 5.8 D, respectively. There is little change for the two values, and they are similar to the Gly result (5.6 D) of Császár.[7] Moreover, it had been demonstrated that polar molecules with dipole moments larger than 2−2.5 D could form stable dipole-bound anions,[65] so ^{1}GlyH1 would be an ideal candidate to be further stabilized by attachment of one or more excess electron(s) with dipole-bound mode as Gly and GlyZW.[36] Both Gly and GlyZW can be the reactants for protonation and can be stabilized by attaching an excess electron. For example, the anionic Gly (^{1}Gly-(−1)) is more stable by 9.0 kcal/ mol than its corresponding neutral form ^{1}Gly0.[36] Moreover, GlyZW can also acts as the reactant of the most stable ^{1}GlyH1, though the spontaneously degenerated form of neutral GlyZW is Gly in the gas phase.[27−36] According to the relationship of ^{1}GlyH1 and the zero-charged glycine isomers and their larger dipole moments, we expect that ^{1}GlyH1 would be further stabilized by attaching excess electron-(s). Then the PA of ^{1}Gly0 would be also greatly affected by the attached excess electron(s). As shown in Figure 1, GlyZW can be stabilized in the gas phase by protonation at its O4 site. Results show the energy of ^{1}GlyH1 decreases by 212.6 kcal/ mol, calibrated at the CCSD(T)/aug-cc-pvdz// B3LYP/6-31++G** level, relative to that of the neutral Gly. In other words, protonation can make GlyZW and Gly more stable. Now we will further probe the stability of the protonated system by

attaching excess electron(s), as Maciej et al.[36] had done for GlyZW and Gly, and calculate the corresponding PAs.

(2) ^{2}GlyH0. As expected, the energy of the ^{2}GlyH0 species reduces by 99.2 and 90.4 kcal/mol, obtained at the B3LYP/6-311++G** and CCSD(T)/aug-cc-pvdz level, respectively, when an excess electron is attached to ^{1}GlyH1 (see Table 1, ^{2}GlyH0). Figure 2 shows that the excess electron mainly locates on three amino hydrogens with 0.43e (H9), 0.29e (H10), and 0.05e (H11) increments, respectively, compared to their counterpart of ^{1}GlyH1. Only 0.28e charge loss occurs on the N3 atom. The sum of 0.49e distribution on the amino group accounts for a 49% proportion of the total excess electron, which indicates that each corresponding geometry parameter of the amino group would change greatly, especially for the N3H9 bond, if the interaction between the electron and the ^{1}GlyH1 is a "covalent" one. In fact, Figure 3 shows that the N3H9, N3H10, and N3H11 bonds are only extended by 0.031, 0.018, and 0.012 Å, respectively. There is not significant change in distance, so the interaction may still be regarded as a dipole-bound one, as had been done in the GlyZW or Gly system.[36] This implies that protonation of GlyZW does not influence the regularity of excess electron localization. On the other hand, the excess electron localization at the amino hydrogens reduces the positive charge population at the corresponding position. Consequently, the dipole moment of ^{2}GlyH0 has been reduced to 1.5 D. Among the three amino hydrogens, H9 is the largest contributor of charge decrease to the whole amino group. The electrostatic interaction between H9 and O5 has greatly decreased, the amino group turns 47.3° around the N3C2 axis with H9 and H10 far away from the O5 site. Besides, as another electron acceptor, the carboxyl group has also to twist 57.0° around the C1C2 axis. As a result, the hydrogen bonding of O5···H11 is extended by 0.714 Å. In addition to the decreased charge populations on the two atoms, the hydrogen-bonding interaction has become relatively weak. However, the geometric deformation can greatly benefit the stability of the excess electron-attached system. Calculations show that the vertical electron affinity (EA_v) is −92.1 and −92.2 kcal/mol, respectively, obtained at the B3LYP level with two different basis sets. At the more accurate level (CCSD(T)), the value is −86.9 kcal/mol. In fact, the relative energy between ^{2}GlyH0 and ^{1}GlyH1 just accounts for the negative adiabatic EA_a. So the deformation energy contribution to the system stability is the difference between the adiabatic EA and the vertical EA. This result shows that the deformation contribution is only 3.5 kcal/mol, far lower than the electronic one. On the other hand, the serious difference of the dipole moments with 1.5 D (EA_a) and 4.5 D (EA_v) can clearly confirm this point. In comparison with the cases of the GlyZW and Gly species attached by an excess electron (the energy drop of Gly is 9.0 kcal/mol[36]), respectively, it can be observed that attachment of an excess electron to ^{1}GlyH1 can more effectively strengthen the stability of the system.

(3) ^{1}GlyH(−1). Upon attaching two electrons to the ^{1}GlyH1 system, another case is observed. The attached electrons still mainly localize at the amino group [2.07e (1.14e)]. In detail, the electrons at the H9, H10, and H11 sites increase by 1.06e (0.46e), 0.69e (0.36e), and 0.73e (0.31e), respectively, while that at the N3 site decreases by 0.41e (−0.11e). Note here that charges in parentheses denote AIM results. $S = 1.14$e (AIM) implies that more than half of two attached electrons mainly localize over the amino group. The tendency is in agreement with that of Mulliken result (2.07e), though the latter overestimates it in absolute value (see Figure 2). Accordingly, the N3H9 and N3H10 are elongated by 0.045 and 0.020 Å, respectively, while the N3H11 keeps constant relative to those of the ^{1}GlyH1 species. This is because the reduction of the electron at the N3 site partly counteracts the electron increase of H11, which results in an almost unchanged N3H11 bond distance (1.046 Å). Due to the increased electron distributions on the amino group and carboxyl group, respectively, the strong electronic repulsion between them makes the amino group turn 58.9° around the C2N3 axis, increasing by 10.7° compared to that of ^{2}GlyH0. Correspondingly, the carboxyl group twists 109.8° around the C1C2 axis. The twist angle increases 52.4° compared to that of ^{2}GlyH0. As a result, the original O5···H11 bond extends to 3.470 Å, basically losing the hydrogen-bonding function. However, the hydrogen bond O5···H6 seems to be strengthened, as evidenced by the shortened distance (2.258 Å) relative to those of ^{1}GlyH1 (2.405 Å) and ^{2}GlyH0 (2.317 Å). Except for the twists of some atom groups induced by the electron(s) attachment, two-electron attachment makes the system almost recover its original magnitude of dipole moment without large changes (7.2 D). The general effect of these factors makes the stability of ^{1}GlyH1 strengthen by 95.8 and 106.2 kcal/mol, respectively, obtained at CCSD(T) and B3LYP levels. Calculated vertical EA_vs are −88.8 and −93.8 kcal/mol at the two different levels, respectively. In contrast to those of ^{2}GlyH0, it can be observed that the decreases of relative energy and EA_v of ^{1}GlyH(−1) are only, respectively, 5.4 and 1.9 kcal/mol, as obtained at the CCSD(T) level. The small EA_v difference indicates that the additional energy drop induced by the attachment of two excess electrons mainly stems from the contribution of the deformation.

(4) ^{2}GlyH(−2), ^{1}GlyH(−3), and ^{2}GlyH2. Different from the cases of ^{1}GlyH1 and ^{2}GlyH0, calculations[62] shows that only three atoms, C1, C2, and N3, hold positive charges in the three-electron-attached ^{1}GlyH1 (^{2}GlyH(−2)), so the corresponding molecular dipole moment decreases to 2.0 D, though ^{2}GlyH(−2) bears more electrons than ^{1}GlyH1 and ^{2}GlyH0. Optimized results show that the dihedral angles H11N3C2C1 and O5C1C2N3 are 49.6° and −35.3°, respectively. The former is only 1.4° larger than the counterpart of ^{2}GlyH0, while the latter is 22.1° lower than the counterpart of ^{2}GlyH0. So both geometries of ^{2}GlyH0 and ^{2}GlyH(−2) have some similarities. However, the stability of ^{2}GlyH(−2) is decreased by 57.6 kcal/mol compared to that of ^{2}GlyH0, but is still strengthened by 32.8 kcal/mol relative to that of ^{1}GlyH1, obtained at the CCSD(T) level. Table 1 shows that the EA_v of ^{2}GlyH(−2) is −23.3 kcal/mol, which indicates that the deformation contribution to the stability is 9.5 kcal/mol, a subordinate one.

As a comparison, the cases of four electrons attached to [^{1}GlyH(−3)] and one electron ionized from ^{1}GlyH1 (^{2}GlyH2) are investigated, respectively. Results show that both geometries of ^{1}GlyH(−3) and ^{2}GlyH2 look like the original ^{1}GlyH1 one. For example, the geometry of ^{1}GlyH(−3) approaches C_s symmetry, while ^{2}GlyH2 is a real C_s-symmetric structure. For ^{2}GlyH(−2) and ^{1}GlyH(−3), the attached electrons in the two systems have partly transferred to the H6 atom, and the electron distributions on the two hydrogens of C_α(C2) also have larger proportion. These indicate that the excess electrons ($n > 2$) begin to transfer from the amino hydrogens to the hydroxyl hydrogen, which results in the energy increase of the corresponding system. Moreover, the deformation contribution of ^{1}GlyH(−3) disfavors its stability. On the other hand, Table S1 shows that the carboxyl oxygen (O5) is the major electron donor (0.47e) when an electron is ionized from ^{1}GlyH1 (see ^{2}GlyH2). Ionization can make the energy of the system increase markedly (384.5 kcal/mol).

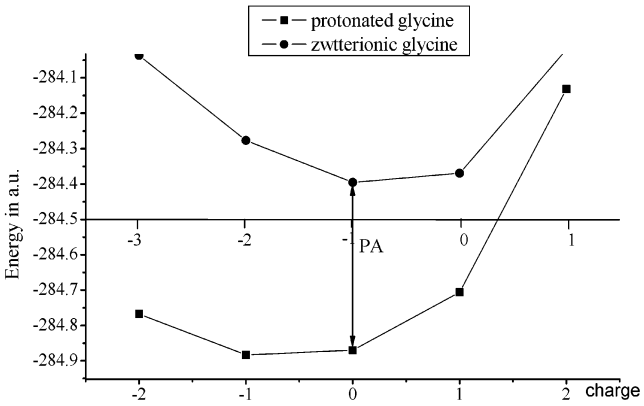

Figure 4. Plots of total energies of charged GlyZW and ^{1}GlyH1 species calculated at the B3LYP/6-31++G** level of theory.

Electronic Effect on the Proton Affinity (PA). Figure 4 displays two PES curves vs different charges, which belong to protonated glycine and the zwtterionic one, respectively. The difference of each corresponding two points in two PESs just corresponds to the PA of the ^{m}GlyZW($n-1$) species. Note that the valence of each ^{m}GlyZW($n-1$) is one less than that of the corresponding ^{m}GlyHn species.

Calculations show that the PA decreases along with the valence increase. For example, the PA only accounts to 71.4 kcal/mol in ^{2}GlyH2, while it is 481.7 kcal/mol in ^{2}GlyH(-2) species, as obtained at B3LYP/6-31++G** level. The CCSD-(T) calibrations for the two values are 68.5 and 473.5 kcal/mol (see Table 1). So the more attached electrons there are, the greater the corresponding PA is, i.e., electron attachment favors the PA. However, more attached electrons disfavors the stability of ^{1}GlyH1 and GlyZW. Table 1 shows that the stability of ^{2}GlyH(-2) has been decreased seriously relative to that of ^{2}GlyH0 and ^{1}GlyH(-1), though it is still more stable than ^{1}GlyH1. So the best estimate or the largest probability for the PA should lie in ^{2}GlyH0 due to two aspects. One is that ^{2}GlyH0 is the second most stable species, with only 5.4 kcal/mol energy promotion than ^{1}GlyH(-1), while ^{2}GlyZW(-1) anion is the most stable one among the different ^{m}GlyZW($n-1$) species. Another is that the energy drop between ^{2}GlyH0 and ^{1}GlyH-(-1) is 7.1 kcal/mol, as obtained at the B3LYP/6-31++G** level, far lower than that between the ^{1}GlyZW(-2) and ^{2}GlyZW(-1) (74.6 kcal/mol) species. In a word, the anionic GlyZW is far more ready to combine with a proton than a neutral species. The PA of the original neural glycine ^{1}Gly0 is 212.6 kcal/mol, as obtained at the CCSD(T) level. The value is in excellent agreement with the recent theoretical (211.1 kcal/mol)[4] and experimental (211.8 kcal/mol)[34] results.

Analyses for the Possible Dissociation Forms of the Series of ^{m}GlyHn Species. Figure 5 displays the possible energies of deprotonation, ionization, affinity of electron(s), and dissociation of hydrogen radical for these ^{m}GlyHn species obtained at the CCSD/aug-cc-pvdz//B3LYP/6-31++G** level. Results show that, obviously, the ^{1}GlyH1 deprotonation will absorb an energy of 212.6 kcal/mol. However, the cases become complicated in other species. For example, ^{2}GlyH2 may capture an electron and then can release 384.5 kcal/mol of energy (EA_a), or it may be divided into ^{2}GlyZW1 and H^+, raising the energy (PA) of the system by 68.5 kcal/mol. So the greatest possibility is that ^{2}GlyH2 can interact with a free electron to form a more stable ^{1}GlyH1 species. For ^{2}GlyH0, it can be separated by following three modes (see **3** in Figure 5). First, it can be deprotonated. However, the needed 316.9 kcal/mol energy (PA) makes the process very difficult. Then, it can ionize an electron and become

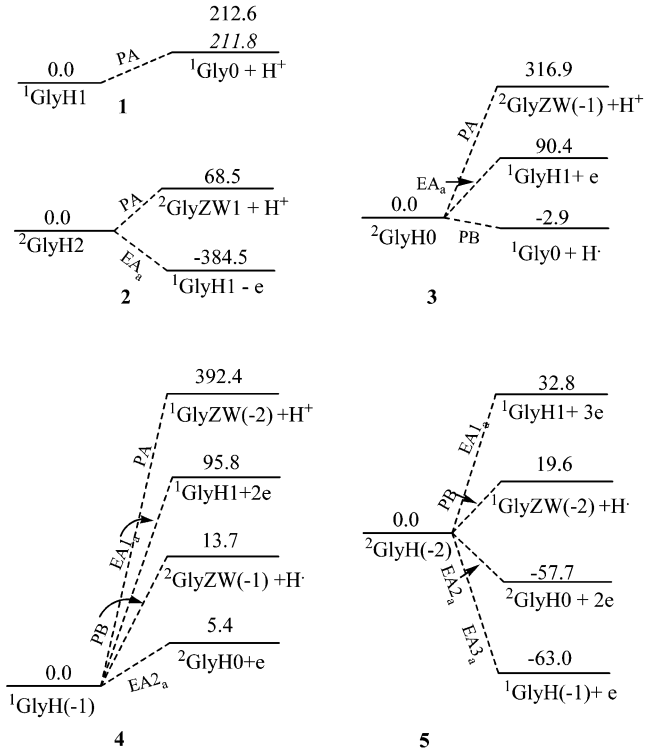

Figure 5. The energies of possible deprotonation (PA), ionization or affinity of electron(s) ($\pm EA_a$), and dissociation of one hydrogen radial (PB) of these ^{m}GlyHn species, obtained at the CCSD/aug-cc-pvdz//B3LYP/6-31++g** level. The result in italic is from ref 35. Energy in kcal/mol.

^{1}GlyH1. The absorption energy of 90.4 kcal/mol ($-EA_a$) makes the process seem more possible relative to the first mode. The last one is to release one hydrogen radical to become the neutral GlyZW (Gly). Calculations show that the energy sum of two separated species is lower by 2.9 kcal/mol in energy than ^{2}GlyH0. So, the third mode becomes the most competitive one. For **4** in Figure 5, four possible dissociation modes are considered. It can be observed that deprotonation of ^{1}GlyH(-1) is almost impossible, due to the too large PA (392.4 kcal/mol), while ionization of two electrons with -95.8 kcal/mol EA_a from the species also seems difficult. Results show that only the fourth mode is the most possible, i.e., ^{1}GlyH(-1) releases an electron to form ^{2}GlyH0, though the ionization energy is -5.4 kcal/mol. In a word, the dissociation of the system with any form would disfavor the stability of the ^{1}GlyH-(-1) system. This conclusion can also be confirmed by the analyses for the case of ^{2}GlyH(-2) dissociation. Results in **5** (see Figure 5) show that loss of three electrons or of a hydrogen radical would make the energy of the system increase. Only loss of one or two electrons can stabilize the system. Especially, loss of one electron from ^{2}GlyH(-2) is the optimal, i.e., ^{1}GlyH-(-1) is the most stable.

Conclusions

A comprehensive DFT (B3LYP) study of the most stable protonated glycine species with various charges in the gas phase was carried out. The relative energies, Pas, and EA_vs were calibrated at the CCSD(T)/aug-cc-pvdz level. The results show that both B3LYP with two different basis sets and CCSD(T) with aug-cc-pvdz basis set offer a consistent ordering for the relative stability and EA_vs of these different charged protonated glycine species. So do for the PA results of these different ^{m}GlyZW($n-1$) species.

Of the most importance is the observation that the combination of protonation and attachment of one or two excess electrons can significantly stabilize the zwitterionic glycine in the gas phase. Moreover, the combination is more effective than either of the two methods singly employed to stabilize GlyZW, as evidenced by the lower relative energy of the electron-attached protonation glycine and larger PA of electron-attached GlyZW. In detail, the energy of ^{1}GlyH1 can be lowered by 90.4 kcal/mol at the CCSD(T) level, when an excess electron is attached. The calculated EA_v is 86.9 kcal/mol at the same level, which indicates that the electronic effect predominates the stability of the system, while the deformation energy is only subordinate (3.5 kcal/mol), though the anionic geometry is twisted seriously. Two-electron attachment can further stabilize ^{1}GlyH1 by 5.4 kcal/mol over ^{2}GlyH0. Calculations show that the additional stability mainly stems from the deformation contribution. Three-electron attached ^{1}GlyH1 is the threshold where the relative energy begins to increase, relative to the one- and two-electron-attached species, though it is stable by 32.8 kcal/mol above ^{1}GlyH1. Either ionizing one electron or attaching four electrons disfavors the stability of ^{1}GlyH1.

Obviously, attachment of more excess electrons would strengthen the proton affinity. However, the one-electron attachment is preferred due to the optimal stability of both ^{2}GlyH0 and ^{2}GlyZW(−1). The PA of ^{2}GlyH0 is 315.4 kcal/mol, far larger than that of ^{1}GlyH1 (212.6 kcal/mol). Note that all the interactions between these attached electrons and ^{1}GlyH1 are dipole-bound ones, as evidenced by little changes in bond distance. Those large twists of geometry under different charge surroundings are only brought on by the need to avoid the larger electrostatic repulsions. All the deformation energies of these protonated glycine species attached by different electron numbers (1, 2, and 3, but not 4) favor the stability of the corresponding system.

The analyses for the possible dissociation forms of the series of ^{m}GlyH*n* species imply that an electron easily attaches to ^{2}GlyH2. The preferred dissociation mode for ^{2}GlyH0 is to be separated into Gly and one hydrogen radical. The possible dissociation cases of both ^{1}GlyH(−1) and ^{2}GlyH(−2) further confirm that both protonation and two-electron combination is the most advantageous for the stability of GlyZW in the gas phase.

Acknowledgment. This work is supported by the National Natural Science Foundation of China (20273040) and the Natural Science Foundation of Shandong Province (Key project/Z2003B01), and the support from SRFDP is also acknowledged.

Supporting Information Available: B3LYP/6-31++G**-calculated Mulliken and AIM charge populations of ^{m}GlyH*n* species. This material is available free of charge via the Internet at http://pubs.acs.org.

References and Notes

(1) Fersht, A. Enzyme Structure and Mechanism, Freeman, W. H.; New York, 1985.

(2) Balaji, V. N.; Ramnarayan, K. *In Biological Active Peptides: Design, Synthesis and Utilization;* Williams, W. V., Weiner, D. B., Eds.; Technomic Publishing: Basel, Switzerland, 1991.

(3) Yu, D.; Armstrong, D. A.; Rauk, A. *Can. J. Chem.* **1992**, *70*, 1762.

(4) Zhang, K.; Chung-Phillips, A. *J. Phys. Chem.* **1998**, *102*, 3625.

(5) Ramek, M.; Cheng, V. K. W.; Frey, R. F.; Newton, S. Q.; Schafer, L. *J. Mol. Struct.* **1991**, *186*, 12, and references therein.

(6) Jensen, J. H.; Gordon, M. S. *J. Am. Chem. Soc.* **1991**, *113*, 7917.

(7) Császár, A. G. *J. Am. Chem. Soc.* **1992**, *114*, 9568.

(8) Frey, R. F.; Conffin, J.; Newton, S. Q.; Ramek, M.; Chang, V. K.; W.; Momany, F. A.; Schafer, L. *J. Am. Chem. Soc.* **1992**, *114*, 5369.

(9) Chipot, C.; Maigret, B.; Rivail, L.-L. *J. Phys. Chem.* **1992**, *96*, 10276.

(10) Vijay, A.; Sathyanarayana, D. N. *J. Phys. Chem.* **1992**, *96*, 10735.

(11) Bliznyuk, A. A.; Schaefer, H. F., III; Amster, I. J. *J. Am. Chem. Soc.* **1993**, *115*, 5149.

(12) Hu, C.-H.; Shen, M.; Schaefer, H. F., III *J. Am. Chem. Soc.* **1993**, *115*, 2923.

(13) Barone, V.; Adamo, C.; Lelj, F. *J. Chem. Phys.* **1995**, *102*, 364.

(14) Reva, I. D.; Plokhotnichenko, A. M.; Stepanian, S. G.; Ivanov, A. Y.; Radchenko, E. D.; Sheina, G. G.; Blagoi, Y. P. *Chem. Phys. Lett.* **1995**, *232*, 141, *Erratum* **1995**, *235*, 617.

(15) Császár, A. G. *J. Mol. Struct.* **1995**, *346*, 141.

(16) (a) Godfrey, P. D.; Brown, R. D. *J. Am. Chem. Soc.* **1995**, *117*, 2019. (b) Godfrey, P. D.; Brown, R. D.; Rodgers, F. M. *J. Mol. Struct.* **1996**, *376*, 65.

(17) Neville, J. J.; Zheng, Y.; Brion, C. E. *J. Am. Chem. Soc.* **1996**, *118*, 10533.

(18) Ramek, M.; Momany, F. A.; Miller, D. M.; Schafer, L. *J. Mol. Struct.* **1996**, *375*, 189.

(19) Sueram, R. D.; Lovas, F. J. *J. Mol. Spectrosc.* **1980**, *72*, 372.

(20) Locke, M. J.; Mclver, R. T. *J. Am. Chem. Soc.* **1983**, *105*, 4226.

(21) Gordon, M. S.; Jensen, J. H. *Acc. Chem. Res.* **1996**, *29*, 536.

(22) Nagy, P. I.; Noszal, B. *J. Phys. Chem. A* **2000**, *104*, 6834.

(23) Ramek, M.; Nagy, P. I. *J. Phys. Chem. A* **2000**, *104*, 6844.

(24) Jensen, F. *J. Am. Chem. Soc.* **1992**, *114*, 9533.

(25) Hoyau, S.; Ohanssian, G. *Chem. Eur. J.* **1998**, *8*, 1561.

(26) Rogalewicz, F.; Ohanessian, G.; Gresh, N. *J. Comput. Chem.* **2000**, *21*, 963.

(27) Ai, H.; Bu, Y.; Han, K. *J. Chem. Phys.* **2003**, *118*, 10973.

(28) Ai, H.; Bu, Y.; Li, P. *Int. J. Quantum Chem.* **2003**, *94*, 205.

(29) Zhang, K.; Zimmerman, D. M.; Chung-Phillips, A.; Cassady, C. J. *J. Am. Chem. Soc.* **1993**, *115*, 10812.

(30) Cassady, C. J.; Carr, S. R.; Zhang, K.; Chung-Phillips, A. *J. Org. Chem.* **1995**, *60*, 1704.

(31) Wright, L. R.; Borkman, R. F.; Gabriell, A. M. *J. Phys. Chem.* **1982**, *86*, 3951.

(32) Bouchonnet, S.; Hoppilliard, Y. *Org. Mass Spectrom.* **1992**, *27*, 71.

(33) Yu, D.; Rauk, A.; Armstrong, D. A. *J. Am. Chem. Soc.* **1995**, *117*, 1789.

(34) Hunter, E. P.; Lias, S. G. *J. Phys. Chem. Ref. Data* In press. See also: Mallard, W. G., Linstrom, P. J., Eds.; NIST Standard Reference Database Number 69, National Institute of Standards and Technology, Gaithersburg, MD, 1997.

(35) Ai, H.; Bu, Y.; Han, K. *J. Chem. Phys.* **2002**, *117*, 7593.

(36) Maciej, G.; Piotr, S.; Jack, S. *J. Am. Chem. Soc.* **2000**, *122*, 10159.

(37) Cantor, C. R.; Schimmel, P. R. *Biophysical Chemistry*; Freeman, W. H., Co.: New York, 1980; Part 1, pp 41−55.

(38) Jensen, J. H.; Gordon, M. S. *J. Am. Chem. Soc.* **1995**, *117*, 8159.

(39) Biemann, K.; Martin, S. A. *Mass Spectrom. Rev.* **1987**, *6*, 1.

(40) Zhang, K.; Chung-Phillips, A. *J. Comput. Chem.* **1998**, *19*, 1862.

(41) Piotr, S.; Janusk, R.; Jack, S.; Maciej, G. *J. Am. Chem. Soc.* **2001**, *123*, 11073.

(42) Aflatooni, K.; Hitt, B.; Gallup, G. A.; Burrow, P. D. *J. Chem. Phys.* **2001**, *115*, 6489.

(43) Li, X.; Cai, Z.; Sevilla, M. D. *J. Phys. Chem.* **2002**, *106*, 1596.

(44) Zhan, C.; Dixon, D. A. *J. Phys. Chem. B* **2003**, *107*, 4403.

(45) (a) Strittmatter, E. F. Lemoff, A. S.; Williams, E. R. *J. Phys. Chem. A* **2000**, *104*, 9793. (b) Bertran, J.; R_Sautiago, L.; Sodupe, M. *J. Phys. Chem. B* **1999**, *103*, 2310. (c) Ai, H.; Bu, Y.; Chen, Z. *J. Chem. Phys.* **2003**, *118*, 1761.

(46) Santiago, L. R.; Sodupe, M.; Oliva, A.; Bertran, J. *J. Phys. Chem. A* **2000**, *104*, 1256.

(47) (a) Becke, A. D. *J. Chem. Phys.* **1993**, *78*, 5648. (b) Petersson, G. A.; Al-Laham, M. A. *J. Chem. Phys.* **1991**, *94*, 6081. (c) Petersson, G. A. Bennett, A.; Tensfeldt, T. G.; Al-Laham, M. A.; Shirley, W. A.; Mantzaris, J. *J. Chem. Phys.* **1988**, *89*, 2193.

(48) (a) Watchers, A. J. H. *J. Chem. Phys.* **1978**, *69*, 3833. (b) Hay, P. J. *J. Chem. Phys.* **1977**, *66*, 4377. (c) Raghavachari, K.; Trucks, G. W. *J. Chem. Phys.* **1989**, *91*, 1062.

(49) Bartlett, R. J. *Annu. Rev. Phys. Chem.* **1981**, *32*, 359.

(50) Raghavachri, K.; Trucks, G. W.; Pople, J. A.; Head-Gordon, M. *Chem. Phys. Lett.* **1989**, *57*, 479.

(51) Kendall, R. A.; Dunning, T. H., Jr.; Harrison, R. J. *J. Chem. Phys.* **1992**, *96*, 6796.

(52) Simon, S.; Sodupe, M.; Bertran, J. *J. Phys. Chem. A.* **2002**, *106*, 5697.

(53) Nguyen, D. T. Scheimer, A. C.; Andzelm, J. W.; Sirois, S.; Salahub, D. R.; Hagler, A. T. *J. Comput. Chem.* **1997**, *18*, 1609.

(54) Gutowski, M.; Skurski, P. *Recent Res. Dev. Phys. Chem.* **1999**, *3*, 245.

(55) (a) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; et al. *Gaussian 94*, Revision D.1, Gaussian, Inc., Pittsburgh, PA, 1994. (b) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; et al. *Gaussian 98*, Revision A.3, Gaussian, Inc., Pittsburgh, PA, 1998.

(56) Messey, V.; Gibson, Q. H.; Veeger, C. *Biochem. J*. **1960**, *77*, 341.

(57) Messey, V.; Veeger, C. *Biochim. Biophys. Acta* **1961**, *48*, 33.

(58) Nishimoto, K. Higashimura, K.; Asada, T. *Theor Chem Acc* **1999**, *102*, 355.

(59) Messey, V.; Gibson, Q. H. *Fed. Proc.* **1964**, *23*, 18.

(60) Searls, B. L.; Peters, J. M.; Sanadi, D. R. *J. Biol. Chem*. **1961**, *236*, 2317.

(61) Eades, R. A.; Scanion, K.; Ellenber, M. R.; Dixon, D. A. *J. Phys. Chem.* **1980**, *84*, 2840.

(62) Supporting Information available. Mulliken and AIM charge (those in the parentheses) distributions of the gly-H-xx system with various charges obtained at the B3LYP/6-31++G** level are listed in Table S1.

(63) Bader, R. F. W. Oxford University Press: Oxford, 1990.

(64) (a) Cioslowski, J.; Nanayakkara, A.; Challacombe, M. *Chem. Phys. Lett.* **1993**, *203*, 137. (b) Cioslowski Surjan, P. R. *J. Mol. Struct.* **1992**, *255*, 9. (c) Cioslowski, J.; Stefanov, B. B. *Mol. Phys*. **1995**, *84*, 707. (d) Stefanov, B. B.; Cioslowski, J. R. *J. Comput. Chem.* **1995**, *16*, 1394. (e) Cioslowski, J. *Int. J. Quantum Chem. Quantum Chem. Symp*. **1990**, *24*, 15. (f) Cioslowski, J.; Mixon, S. T. *J. Am. Chem. Soc.* **1991**, *113*, 4142. (g) Cioslowski, J. *Chem. Phys. Lett.* **1992**, *194*, 73. (h) Cioslowski, J. *Chem. Phys. Lett*. **1994**, *219*, 151.

(65) Desfrancois, C.; Abdoul-Carime, H.; Khelifa, N.; Scherman, J. P.; Bremmer, V.; Millie, P. *J. Chem. Phys.* **1995**, *102*, 4952.