EH stabilization energy is ca. −11 eV. Once again the CNDO/2 and INDO minima occur in regions of high overlap and the inclusion of one-center exchange integrals serves only to increase the stabilization energy. Similar CNDO/2 and INDO calculations on the approach of two eclipsed molecules of ammonia led to weakly repulsive potential curves, whereas EH calculations led to strongly repulsive curves.

Concluding Remarks

The model calculations reported here show that the popular CNDO/2 and INDO CS RHF methods impart too much stability to systems containing nitrogen and/or oxygen atoms separated from one another or from π bonds by less than 4 Å. As the separation in question decreases and the AO overlap increases, the total electronic energy is further underestimated with the result that the potential energy surface becomes increasingly unrealistic. These observations should be useful for assessing the significance of many of the CNDO/2 and INDO calculations that are being reported in growing numbers alongside experimental results.

Our results provide unequivocal evidence that one cannot be sure at the outset of a project that a semiempirical MO theory will give results that are qualitatively correct or that a more sophisticated theory will generally give better results.[11] However, assumptions of this type still have wide currency. One should establish their validity each time a new application of a theory is envisaged, either by careful reference to the literature or by a preliminary series of carefully designed numerical tests. The direct σ-type interaction between two lone pairs is a very simple, but conclusive test of theory that has with few exceptions[12] been overlooked.

Finally, it should be noted that our calculations complement recent work by other authors. Veillard[13] has reviewed the literature and established that the CNDO/2 method fails to predict correctly the conformational preferences of molecules $H_mA\text{-}BH_n$ and their substituted derivatives, when both atoms A and B bear one or two lone pairs. And Giessner-Prette and Pullman[14] have concluded from the calculation of molecular electrostatic potentials that the CNDO method makes oxygen too attractive (as a site for protonation) compared to nitrogen.

Acknowledgment. We thank I. G. Ross for stimulating discussions and Leo Radom for performing some ab initio calculations on the water/ethylene system. One of us, A.R.G., is also indebted to the Australian Government for a Commonwealth Post-Graduate Award and to the Australian National University for a Research Scholarship.

References and Notes

(1) Present address: Department of Chemistry, New South Wales Institute of Technology, P. O. Box 123, Broadway, N.S.W., Australia 2007.
(2) J. A. Pople, and G. A. Segal, *J. Chem. Phys.*, **44**, 3289 (1966); J. A. Pople, D. L. Beveridge, and P. A. Dobosh, *ibid.*, **47**, 2026 (1967); J. A. Pople and D. L. Beveridge, "Approximate Molecular Orbital Theory", McGraw-Hill, New York, N.Y., 1970.
(3) (a) A. R. Gregory, Ph.D. thesis, The Australian National University, 1972, and references cited therein; (b) A. R. Gregory, *Jerusalem Symp. Quantum Chem. Biochem.*, **6**, 23 (1974).
(4) P. A. Kollman and L. C. Allen, *Chem. Rev.*, **72**, 283 (1972), and references cited therein.
(5) A. R. Gregory and M. N. Paddon-Row, unpublished work.
(6) The program that was used is a modified version of Program No. 141, "CNINDO", distributed by the Quantum Chemistry Program Exchange (QCPE), Indiana University, Bloomington, Ind.
(7) R. Hoffmann, *J. Chem. Phys.*, **39**, 1397 (1963).
(8) The program that was used is a modified version of Program No. 64, "EXTHUC", distributed by QCPE.
(9) R. Hoffmann, S. Swaminathan, B. G. Odell, and R. Gleiter, *J. Am. Chem. Soc.*, **92**, 7091 (1970).
(10) L. Radom, private communication.
(11) R. B. Davidson, W. L. Jorgensen, and L. C. Allen, *J. Am. Chem. Soc.*, **92**, 749 (1970); P. A. Kollman and L. C. Allen, *Ibid.*, **92**, 753 (1970).
(12) W. Adam, A. Grimison, R. Hoffmann, and C. Z. de Ortiz, *J. Am. Chem. Soc.*, **90**, 1509 (1968).
(13) A. Veillard, *Chem. Phys. Lett.*, **33**, 15 (1975).
(14) C. Giessner-Prette and A. Pullman, *Theor. Chim. Acta*, **37**, 335 (1975).

Theoretical Study of the Protonation of Glycine in Gas-Phase Ion–Molecule Reactions

K. Chung, R. M. Hedges,* and R. D. Macfarlane*

Contribution form the Cyclotron Institute and Department of Chemistry,
Texas A&M University, College Station, Texas 77843.

Received September 18, 1975

Abstract: The electronic structure of glycine and its ion–molecule association reaction with H^+ have been studied using a semiempirical INDO-type molecular orbital computation. The results indicate that the association reaction of glycine with H^+ produces a quasi-molecular ion possessing two functional groups, NH_2 and COOH, interacting through N···HO type hydrogen bonding. However, a wide range of conformations is suggested due to the low energy barriers among the several different configurations.

The low yield of parent ion of amino acids produced by electron impact has prompted the mass spectrometric studies of the biological molecules with more gentle means of ionization. Examples include chemical ionization,[1] field desorption,[2] and ^{252}Cf-Plasma desorption,[3] which have been found to form relatively stable products by the ion–molecule association reaction,

$$M + A^+ \rightarrow MA^+$$

$$MA^+ \rightarrow \text{fragments}$$

Among quasi-molecular (QM) ion products, the MH^+ formed by protonation appears as a major component for most ion–molecule association reactions occurring in mass spectrometry. To understand the condition and mechanisms of the ion production by this method, theoretical investigation of the interaction of the simplest α-amino acid, glycine, with free H^+ ions was undertaken using semiempirical INDO molecular orbital theory.[4]

Glycine is known to exist in a variety of structures depending on its environment. In solution, it exists either as the zwitterion,

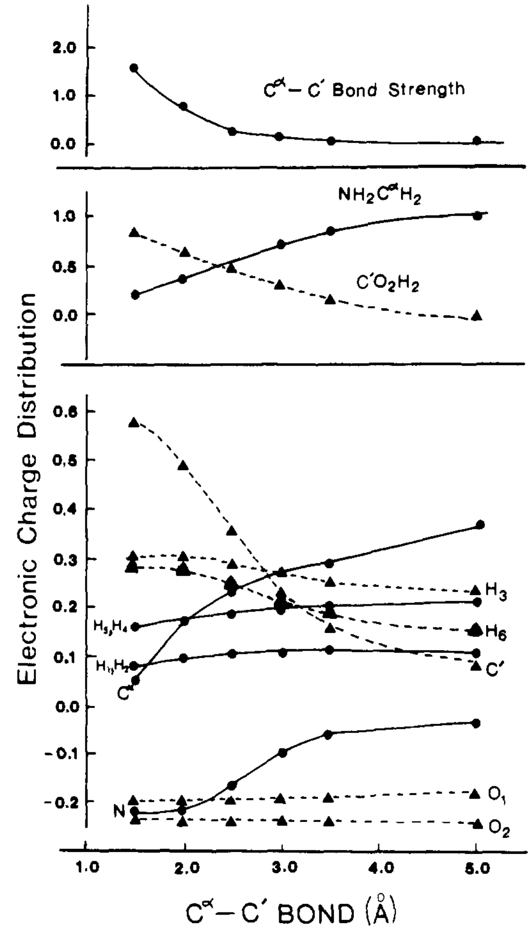
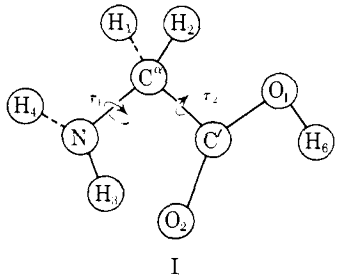


Figure 1. Electronic charge variation in the atoms of QM^+ vs. C^α-C' bond separation.

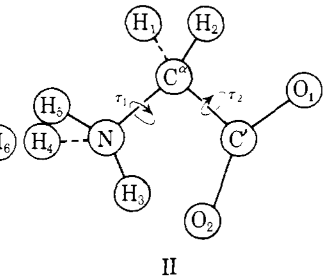
$H_3N^+CH_2COO^-$, or glycinum ion, $NH_3^+CH_2COOH$, depending on the pH of the solvent, while in the isolated state it is suggested to take the neutral form, H_2NCH_2COOH.[5] X-ray diffraction studies on glycine have shown that all of the heavy atoms in the glycinium ions lie in a plane,[6] while in the zwitterion slight displacement has been observed.[6,7] In the present study, therefore, several possible conformations of glycines were undertaken and the computational result of glycines with H^+ ions is compared with the experimental data.

Results with Computation

Structure of Glycine and Its Zwitterion. Since the heavy atoms in glycine (I) and its zwitterion (II) lie almost in a plane, the calculations were undertaken first assuming all the heavy atoms (C^α, C', N, O_1, and O_2) and some hydrogen atoms (H_3 and H_6) lie in a plane. The optimum conformation for glycine was found by rotating the functional groups, NH_2 and COOH of glycine (I) and the NH_3^+ and COO^- groups of the zwitterion (II), by the angle of (τ_1,τ_2). Geometrical parameters were fixed at Pople's values.[4]



I



II

Table I. Bond Lengths, Energies, and Population Analyses of Glycine and Its Protonated Ions

	Glycine		Glycine-H^+		
	I	II	I-H^+	II-H^+	QM^+
			Bond Length, Å		
C'-O_1	1.43	1.30	1.43	1.30	1.30
C'-O_2	1.22	1.30	1.22	1.30	1.30
O_2-H_3		1.98		1.98	1.07
N-H_3		1.00	1.04	1.00	1.56
			Total Energy, au		
	−63.7698	−63.6635	−64.2846	−64.2988	−64.3202
			Population Analyses (Mulliken Type)		
C^α	0.090	0.037	0.055	0.053	0.048
C'	0.0464	0.412	0.463	0.475	0.575
O_1	−0.333	−0.519	−0.290	−0.249	−0.203
O_2	−0.353	−0.577	−0.316	−0.379	−0.229
N	−0.210	0.064	0.062	0.050	−0.217
C^α-C'	1.290	1.196	1.560	1.560	1.590
C^α-N	1.580	1.496	1.227	1.232	1.278
C'-O_1	1.107	1.532	1.128	1.416	1.481
C'-O_2	1.825	1.520	1.828	1.587	1.486
N-H_3		1.180	1.178	1.162	0.216
N-H_4	1.191	1.180	1.178	0.210	1.190
N-H_5	1.191	1.162	1.178	0.210	1.190
O_1-H_6	1.001		1.003	0.992	1.008
O_2-H_3		0.023	0.000	0.033	0.766

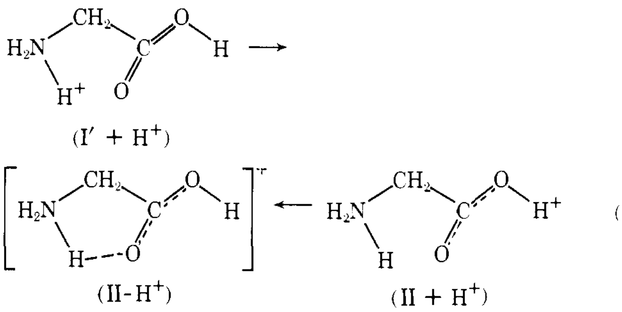
The conformational energy surfaces calculated in 10° increments of τ_1 and τ_2 in the isolated state showed the absolute minimum at the angles of (57,−20) and (0,0) for glycine (I) and its zwitterion (II). The computed results are summarized in Table I for molecular energies and population analyses. Dipole moments of the neutral glycine (I) and the zwitterion form (II) have also been determined in the present study for the minimum energy conformations. They have the magnitude of 2.38 and 13.37 D for conformations I and II, respectively. The dipole movement of the zwitterion is in excellent agreement with the experimental value of 13.3 D in aqueous solution as measured by Buckingham.[8]

The charge distributions in Table I are in good agreement with the previously reported values of Del Re and Pullman[9] using extended Huckel procedures and those of Oegerle and Sabin[10] with the CNDO method. Ryan and Whitten[11] also have calculated the electronic structure of the zwitterion form of glycine using ab initio SCF computation. The charge distributed on each atom was rather different from the present results. But the total charge distributed on functional groups is in quite good agreement as:

NH_3^+—	CH_2—	COO^-	
0.57	0.15	−0.72	Ryan and Whitten (ab initio)[11]
0.60	0.09	−0.69	Present study (INDO)

Shipman and Christoffersen[12] and Ryan and Whitten[11] have obtained the same ordering of the valence molecular orbitals of the zwitterion form of glycine using different types of ab initio computation. The characters of the corresponding orbital energies using the INDO method are also found to be comparable with those obtained with ab initio type methods. Thus, further calculations based on these structures were continued.

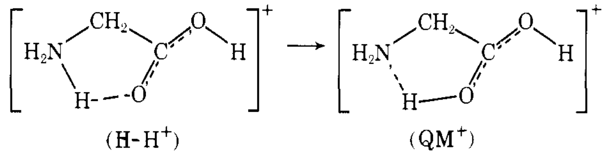
Reaction of Glycine with H^+. The computational results for the association of a H^+ to glycine via several possible routes indicate that the proton approaches the oxygen atom of the zwitterion from the main molecular plane (II-H^+), and approaches the nitrogen of neutral glycine along the direction of the nitrogen's electron lone pair (I-H^+). This protonated glycine (I-H$^+$) is stabilized further by rotating the NH_3^+ group by 60°. Since the rotational barrier of the amino group is relatively small (<2 kcal) compared to the glycine-H$^+$ association energy (~300 kcal), the approach of the H$^+$ to glycine was studied assuming the rotation of the amino group is followed by the attachment of proton. Thus, the energetics were investigated when the lone pair electrons of nitrogen of neutral glycine (I) lie in the main molecular plane by rotating the NH_2 group. For both incidents, the approach is most favored by producing a QM ion of glycine in the forms shown in eq 1,



(1)

which are similar in that they each possess one hydrogen atom between the NH_2 and COOH groups and, consequently, form a planar ring-type structure. However, a wide range of conformations is suggested due to the low energy barriers among the several different configurations.

Hydrogen (H_3) Transfer in QM Ions of Glycines (II-H$^+$). Although there is no direct experimental evidence for the rearrangement of the QM ions in the reaction system, products are observed that are formed by a hydrogen transfer in chemical ionization followed by the cleavage of the C$^\alpha$–C′ bond and the loss of $COOH_2$ (other possible molecules such as CO_2, CO, H_2, H_2O, etc.). The residual fragment ion, $NH_2CH_2^+$, appears as a strong mass peak in addition to the QM ions.[1-3] According to our present results, H_3 transfer could be expected to occur most efficiently through [$NH_3 \cdots O_2 \rightarrow N \cdots H_3O_2$] type



hydrogen transfer because of the largest negative charge of O_2 in the QM ions as well as the planar ring-type hydrogen bonding conformation of the ions. In the hope of calculating the energy barrier for the hydrogen transfer, the energy surfaces for the movement of the hydrogen (H_3) between N and O_2 were computed. This contour has a rather interesting character with only one minimum favoring N···HO type ring structure (QM$^+$). The QM ion stabilized by a hydrogen transfer (QM$^+$) from the NH_3 group to the COOH group will have not much difficulty in producing the $NH_2CH_2^+$ as:

$$\mathrm{QM}^+ \xrightarrow{\mathrm{C}^\alpha\text{–}\mathrm{C}'\text{ cleavage}} \mathrm{NH_2CH_2}^+ + \mathrm{COOH_2}\ \text{(other products)}$$

Discussion and Summary

The computational results for the protonation of glycine in the isolated state show that the proton approaches the lone pair electrons of nitrogen (I) or oxygen atoms (II) of the glycine molecule. The presence of several local minima in the contour maps with low energy barriers among the minima suggest the possibility of multiconformation of the QM ions in the reaction system. The QM ions thus formed are further stabilized by forming a planar ring-type structure through N···HO_2 type hydrogen bonding (QM$^+$).

The so-called stabilization of the QM ions seems to occur partly by increasing the π-bond character around the heavy atoms. The delocalized π-bond character around the heavy atoms appears only for II-H$^+$ and QM$^+$, which possess the planar ring-type structure through the N···H···O hydrogen bonding. Further stabilization of the QM ions seems to occur through redistributing the nonbonding characters localized mainly in oxygen atoms in case of NH···O type QM ion (II-H$^+$) among the three electronegative atoms, N, O_1, and O_2 (QM$^+$). Such redistribution is obtained through hydrogen transfer from the nitrogen to oxygen atom of the protonated glycine as [($NH_3 \cdots O_2$) → ($N \cdots H_3O_2$)]. The QM ions, now possessing $H_2NC^\alpha H_2C'(OH)_2$ type structure, will not have much difficulty in producing the $NH_2CH_2^+$ fragment ion through C$^\alpha$–C′ bond cleavage.

The variation of charge distribution on the atoms along the C$^\alpha$–C′ bond scission of the QM ion (QM$^+$) is illustrated in Figure 1. The fragmentation processes seem accompanied by an extensive charge rearrangement. Before bond cleavage, most of the positive charge is carried by the $COOH_2$ group rather than $NH_2CH_2^+$ group. However, as the C$^\alpha$–C′ bond is extended, the major positive charge carried by C′ atom in the $C'OOH_2$ group is now transferred to C$^\alpha$ and N atoms of the $NH_2CH_2^+$ group and, finally, the positive charge becomes delocalized in the atoms of the $NH_2CH_2^+$ group.

Although the computational results on the glycine itself with the INDO method are in general found to be in good agreement with the previously reported ab initio results, the semiempirical equilibrium geometries of the protonated glycines may fail to represent true reaction schemes, as observed in a small molecular system by favoring anomalously small bond angles.[13] However, we hoped that, for large molecules like glycine and its protonated ions, the present results may not be much off from the actual reaction pathways or the true equilibrium geometries of the products. Further work along these lines (nevertheless) should be done with more accurate computational methods.

Acknowledgment. This work has been supported in part by the U.S. ERDA and Grants A-106 and A-258 of the Robert A. Welch Foundation.

References and Notes

(1) P. A. Leclercq and D. M. Desiderio, *Org. Mass Spectrom.*, **7**, 515 (1973); G. W. A. Milne, T. Axenrod, and H. M. Fiales, *J. Am. Chem. Soc.*, **92**, 5170 (1970).
(2) H. U. Winkler and H. D. Beckey, *Org. Mass Spectrom.*, **6**, 655 (1972).
(3) D. F. Torgerson, R. P. Skowronski, and R. D. Macfarlane, *Biochem. Biophys. Res. Commun.*, **60**, 616 (1974).
(4) J. A. Pople and D. L. Beveridge, "Approximate Molecular Orbital Theory", McGraw-Hill, New York, N.Y., 1970.
(5) K. Biemann, J. Seibl, and F. Gapp, *J. Am. Chem. Soc.*, **83**, 3795 (1961); G. Junk and H. Svee, *ibid.*, **85**, 839 (1963).
(6) S. Hoshino, Y. Okaya, and R. Repinsky, *Phys. Rev.*, **115**, 323 (1959).
(7) P.-G. Jonsson and A. Kvick, *Acta Crystallogr., Sect. B*, **28**, 1827 (1972).
(8) A. D. Buckingham, *Aust. J. Chem.*, **6**, 323 (1953).
(9) G. Del Re, B. Pullman, and T. Yonezawa, *Biochim. Biophys. Acta*, **75**, 153 (1963).
(10) W. R. Oegerle and J. R. Sabin, *J. Mol. Struct.*, **15**, 131 (1973).
(11) J. A. Ryan and J. L. Whitten, *J. Am. Chem. Soc.*, **94**, 2396 (1972).
(12) L. L. Shipman and R. E. Christoffersen, *Theor. Chim. Acta*, **31**, 75–82 (1972).
(13) W. R. Wadt and W. A. Goddard, III, *J. Am. Chem. Soc.*, **96**, 1689 (1974).