Collision Energy Dependence of Proton-Bound Dimer Dissociation: Entropy Effects, Proton Affinities, and Intramolecular Hydrogen-Bonding in Protonated Peptides

Xueheng Cheng, Zhuchun Wu, and Catherine Fenselau*

Contribution from the Department of Chemistry and Biochemistry, University of Maryland Baltimore County, Baltimore, Maryland 21228

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Abstract: The ratios of product ions from dissociation of proton-bound dimers have been measured at various collision energies using a four-sector tandem mass spectrometer. Dissociation of proton-bound dimers $[\mathrm{BuNH_2} + \mathrm{Me_2NH}]\mathrm{H}^+$, $[\text{c-}\mathrm{C_6H_{11}NH_2} + \mathrm{Me_2NH}]\mathrm{H}^+$ and $[\mathrm{Gly_3} + \mathrm{Gly_4}]\mathrm{H}^+$ showed collision energy dependence consistent with little or no entropy effects. In contrast, dissociation of proton-bound dimers $[\mathrm{B_i}$ + 1,4-butanediamine$]\mathrm{H}^+$, $[\mathrm{B_i} + \mathrm{Gly_3}]\mathrm{H}^+$, and $[\mathrm{B_i} + \mathrm{Gly_4}]\mathrm{H}^+$ ($\mathrm{B_i}$ = a series of monoamines with known proton affinity (PA)) showed pronounced entropy effects. Using the assumption that the ratio of the partition functions for the activated complexes leading to the two competing dissociation products, $Q_i^\ddagger/Q^\ddagger$, is constant in the range of the dimer internal energies studied, the values of PA and R ln $(Q_i^\ddagger/Q^\ddagger)$ were obtained by linear regression. These values for 1,4-butanediamine agree well with the literature PA and with $\Delta(\Delta S^\circ_{\mathrm{H}^+})$, the difference in the entropy of protonation between monoamines and 1,4-butanediamine. Proton affinities for $\mathrm{Gly_3}$ and $\mathrm{Gly_4}$ obtained using this procedure are higher by 3–5 kcal/mol than those reported earlier using the standard kinetic method. The entropy effects in $\mathrm{Gly_3}$ and $\mathrm{Gly_4}$ suggest the existence of intramolecular hydrogen-bonding in these protonated peptides. This method of examining entropy effects in proton-bound dimer dissociation should be feasible for other systems and useful for the investigation of thermochemical properties and conformations of gas-phase ions of complicated structure.

Introduction

The study of physical properties[1] and chemical reactivities[2] of biopolymer ions in the gas phase has become an active research field due to the progress in the techniques for the generation of these ions.[3] Research in this field is important, in a fundamental way, to our understanding of the behavior of these large ions in a solvent-free environment and, in the applied aspect, to the structural determination of biopolymers using mass spectrometry. Proton affinity (PA)[4] is one of the basic properties of molecules in the gas phase. Relative PA is usually determined by measuring relative gas-phase basicity (GB) using the equilibrium method[5] at one temperature, and the entropy of protonation ($\Delta S^\circ_{\mathrm{H}^+}$), which is required to calculate the PA, is usually estimated using statistical thermodynamics.[6] Relative PA can also be determined directly using variable-temperature equilibrium methods.[7] However, for bipolymers, the low volatility limits the use of equilibrium approaches. A few recent studies on the PA of amino acids,[1a–d] peptides,[1e] and nucleosides[1f] employed either the bracketing method,[8] in which the positive identification of a reaction is equated to the exothermicity or exoergodicity of the process and vice versa, or the kinetic method,[9] in which the relative PA is obtained from the ratios of the product ion abundance in the dissociation of proton-bound dimers.

The kinetic method, developed by Cooks and co-workers,[9] uses a kinetic measurement (relative rate of dissociation of a proton-bound dimer to each of the individual protonated bases) to approximate the thermochemical property (relative PA of the two bases which form the proton-bound dimer). For the dissociation of proton-bound dimers formed between a series of reference bases $\mathrm{B_i}$ and the base B whose PA is to be determined

$$\begin{tikzcd}[column sep=small, row sep=small] & \mathrm{B} + \mathrm{B_iH^+} \\ {[\mathrm{B_i} + \mathrm{B}]\mathrm{H}^+} \arrow[ur, "k_i"] \arrow[dr, "k"'] & \\ & \mathrm{B_i} + \mathrm{BH^+} \end{tikzcd} \qquad (1)$$

The ratio of rate constants can be derived using unimolecular

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[4] For the process of protonation of a base B in the gas phase, $\mathrm{B} + \mathrm{H}^+ \rightarrow \mathrm{BH}^+$, the proton affinity of B, PA(B), is defined as the negative of the enthalpy change and the gas-phase basicity of B, GB(B), is defined as the negative of the free energy change. The entropy change, entropy of protonation, is presented as $\Delta S^\circ_{\mathrm{H}^+}$ in this paper. For more information and compilation of PA and GB, see: (a) Lias, S. G.; Liebman, J. F.; Levin, R. D. *J. Phys. Chem. Ref. Data* **1984**, *13*, 695–808. (b) Lias, S. G.; Bartmess, J. E.; Liebman, J. F.; Holmes, J. L.; Levin, R. D. Mallard, W. G. *J. Phys. Chem. Ref. Data, Suppl.* **1988**, *17*, 1–872.

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reaction theory[10]

$$\ln(k_i/k) = \ln(Q_i^{\ddagger}/Q^{\ddagger}) + (\epsilon_0 - \epsilon_0(i)]/RT \qquad (2)$$

where $Q_i^{\ddagger}$ and $Q^{\ddagger}$ are the partition functions of the activated complex for the formation of $B + B_iH^+$ and of $B_i + BH^+$, respectively, $\epsilon_0(i)$ and ϵ_0 are the corresponding activation energies for dissociation, and T is the effective temperature of the dimer ion undergoing dissociation. With the assumption that no reverse barriers exist,[9] the difference in activation energies can be replaced by the difference in PAs

$$\ln(k_i/k) = [\ln(Q_i^{\ddagger}/Q^{\ddagger}) - \mathrm{PA(B)}/RT] + \mathrm{PA(B_i)}/RT \qquad (3)$$

With a second assumption that $Q_i^{\ddagger} \approx Q^{\ddagger}$, which should hold when B_i and B are structurally similar bases,[9] $\ln(Q_i^{\ddagger}/Q^{\ddagger}) \approx 0$ and eq 3 simplifies to

$$\ln(k_i/k) = -\mathrm{PA(B)}/RT + \mathrm{PA(B_i)}/RT \qquad (4)$$

Plotting $\ln(k_i/k)$ vs $\mathrm{PA(B_i)}$ will give the effective temperature and PA(B).

However, when applying the kinetic method to biomolecules such as peptides, it is difficult to find appropriate reference bases that are structurally similar to, and with PA close to, the peptides. If a series of reference bases are used that are structurally dissimilar from the peptides but similar among themselves, the term $\ln(Q_i^{\ddagger}/Q^{\ddagger})$ in eq 3 may not be negligible, but plotting $\ln(k_i/k)$ vs $\mathrm{PA(B_i)}$ will still give a straight line due to the fact that $\ln(Q_i^{\ddagger}/Q^{\ddagger})$ is likely constant among B_i. The value of apparent PA obtained by this treatment, however, is $\mathrm{PA(B)} - RT\ln(Q_i^{\ddagger}/Q^{\ddagger})$. Notice that with the assumption of no reverse barrier, the second term here is equal to $T\Delta(\Delta S^\circ_{H^+})$ with the term $\Delta(\Delta S^\circ_{H^+})$ being the difference in entropy of protonation between the reference bases B_i and the base B whose PA is to be determined. Therefore, the result in such a case is more appropriately presented as gas-phase basicity of B, GB(B), instead of PA(B).

Nevertheless, the knowledge of PA and $\Delta S^\circ_{H^+}$ is very desirable since they signify electronic and structural effects and could be very important for correlating thermochemical properties with the three-dimensional structures of peptides in the gas phase. Use of the statistical thermodynamics approach to estimate the $\Delta S^\circ_{H^+}$ is very difficult here because it requires assumptions about the structure of the ions including the conformation and the sites of protonation which are not clear at this stage.[11]

In this paper, we present an experimental approach to examine the effect of entropy on proton-bound dimer dissociation and to identify proton-bound dimer systems that show pronounced entropy effects. With the assumption that the ratio of the partition functions for the activated complexes leading to the two competing dissociation products, $Q_i^{\ddagger}/Q^{\ddagger}$, is constant in the range of the dimer internal energy studied, the values of PA and $\Delta(\Delta S^\circ_{H^+})$ can be estimated. The method is based on the study of product ion abundance ratios in collision-induced dissociation (CID) of proton-bound dimers at various collision energies.

The effective temperature in eq 3 represents the degree of excitation of the proton-bound dimers undergoing dissociation. It has been demonstrated[12] that the effective temperature increases with the increase in collision energy and the pressure of the target gas in the CID process, which is consistent with the notion that both will increase the internal excitation of the dimer ions. At effective temperature T_j, eq 3 can be written as

$$\ln[k_i(T_j)/k(T_j)] = [\ln[Q_i^{\ddagger}(T_j)/Q^{\ddagger}(T_j)] - \mathrm{PA(B)}/RT_j] + \mathrm{PA(B_i)}/RT_j \qquad (5)$$

Assuming the ratio of $Q_i^{\ddagger}/Q^{\ddagger}$ is constant in the range of effective temperatures studied, plotting $[\ln(Q_i^{\ddagger}/Q^{\ddagger}) - \mathrm{PA(B)}/RT_j]$ vs $1/RT_j$ will give PA(B) and $\ln(Q_i^{\ddagger}/Q^{\ddagger})$ and $\Delta(\Delta S^\circ_{H^+}) = R\ln(Q_i^{\ddagger}/Q^{\ddagger})$ with zero-reverse-barrier assumption.

Experimental Section

A JEOL (Tokyo, Japan) HX110/HX100 four-sector tandem mass spectrometer ($E_1B_1E_2B_2$ geometry) was used in this work. The interface connecting MS1 (E_1B_1) and MS2 (E_2B_2) in the third field-free region (FFR) has been modified to improve ion transmission at low kinetic energies.[13]

Proton-bound dimer ions were generated using fast-atom bombardment (FAB) ionization (glycerol, thioglycerol, or 2-nitrobenzylalcohol matrix, 5 keV Xe atoms). In the mass-analyzed ion kinetic energy (MIKE)[14] experiments, the dimer ion was accelerated to 10 keV kinetic energy and mass-selected using MS1, and the products from unimolecular dissociation in the third FFR were detected by the second electrostatic analyzer (E_2). In the CID experiments, the dimer ion was accelerated to 10 keV (V_{acc}) kinetic energy, mass-selected using MS1, decelerated by three cylindrical lenses, and injected into the collision cell located in the third FFR, where the dimer ion collides with added target gases (He in this work). The potential of the collision cell was set to V_{coll} to achieve the desired collision energy $E_{lab} = e(V_{acc} - V_{coll})$. The product and surviving precursor ions were reaccelerated to 10 keV and detected by a linked-scan at constant B_2/E_2 when $E_{lab} > 100$ eV or by scanning the magnetic field in MS2 (B_2) when $E_{lab} \leq 100$ eV. Under the experimental conditions used in this work, the settings of E_2 slits had an energy band-pass of Ca. ± 110 eV. Thus, scanning the magnetic field in MS2 (B_2) will detect all the product ions with kinetic energy equal to that of the precursor ±110 eV without mass discrimination. The pressure of the target gas He was adjusted to give 30% attenuation of the dimer ion. In the MIKE and CID spectra, ions corresponding to protonated species of one or both of the bases that form the dimer were generally the major products. Minor amounts of ions were sometimes observed corresponding to elimination of 17 and 18 Da from the dimer ion. The ion abundance ratios in the MIKE experiments were calculated from the areas of the peaks, and those in the CID experiments, from the heights of the peaks. Each spectrum was the average of three to five scans, and the uncertainty in the reproducibility of the MIKE experiments was no more than 10% and that of the CID experiments was 10–20%.

All the chemicals were from commercial suppliers and were used as receiveed. For the FAB generation of the proton-bound dimer ions, the two bases were dissolved in 0.1% trifluoroacetic acid (TFA) (1.0% HCl or AcOH added if necessary) and 2 μL of solution was applied to the FAB probe. The ratios of the two bases and the FAB matrix were varied to obtain optimal abundances of the proton-bound dimer ions.

Results and Discussion

We studied the product ion ratios from spontaneous dissociation of several proton-bound dimer systems using the MIKE method and, at various collision energies, using the CID method. The reference bases used and their PAs are listed in Table I. These are all monoamines having similar $\Delta S^\circ_{H^+}$ values ($-\Delta S^\circ_{H^+} = 25.8$–$27.2$ eu from experimental determination or from statistical thermodynamics estimation[4]). The results are shown in Figures 1–5, where the logarithms of the product ion ratios are plotted as a function of collision energy in the center-of-mass frame (E_{cm}). The points for $E_{cm} = 0$ eV are from the MIKE experiments. For proton-bound dimers in which the two bases are expected to have similar $\Delta S^\circ_{H^+}$ values, $[c\text{-}C_6H_{11}NH_2 + Me_2NH]H^+$ and $[BuNH_2 + Me_2NH]H^+$, the logarithms of the product ion ratios for both pairs approach zero as the collision energy is increased (Figure 1), just as expected from eq 4. Similar observations have been made before.[12a,15] In contrast, markedly different behaviors were

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Table I. Reference Bases Used in the Dissociation of Proton-Bound Dimers[a]

1,4-butanediamine		Gly_3		Gly_4	
B_i	PA (kcal/mol)	B_i	PA (kcal/mol)	B_i	PA (kcal/mol)
Bu_3N	234.8	Me_3N	225.1	Bu_2NH	228.4
Pr_3N	234.0	$PhNMe_2$	223.4	Pr_2NH	227.5
Et_3N	232.3	*tert*-amylamine	222.3	Et_2NH	225.9
sec-Bu_2NH	231.8	c-$C_6H_{11}NH_2$	221.2	Me_3N	225.1
Et_2NMe	230.0	Me_2NH	220.6	$PhNMe_2$	223.4
		$BuNH_2$	218.4	*tert*-amylamine	222.3

[a] PA values are from ref 4.

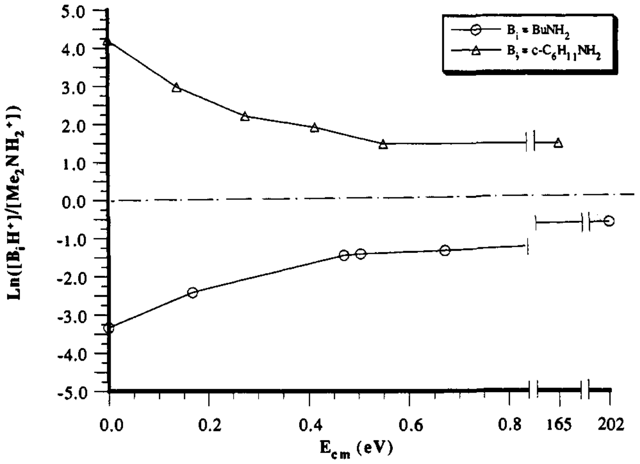

Figure 1. Plot of the logarithms of the ratios of product ions from dissociation of proton-bound dimers containing Me_2NH and monoamines against collision energy in the center-of-mass frame.

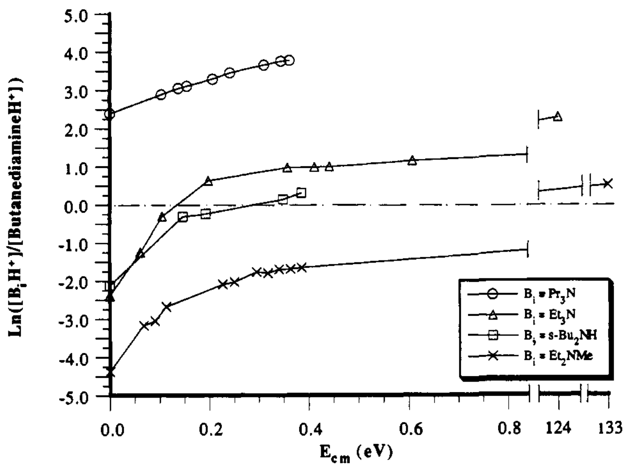

Figure 2. Plot of the logarithms of the ratios of product ions from dissociation of proton-bound dimers containing 1,4-butanediamine and monoamines against collision energy in the center-of-mass frame.

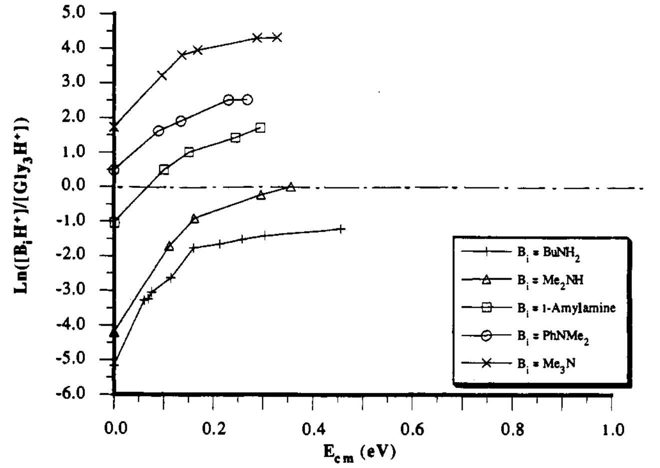

Figure 3. Plot of the logarithms of the ratios of product ions from dissociation of proton-bound dimers containing Gly_3 and monoamines against collision energy in the center-of-mass frame.

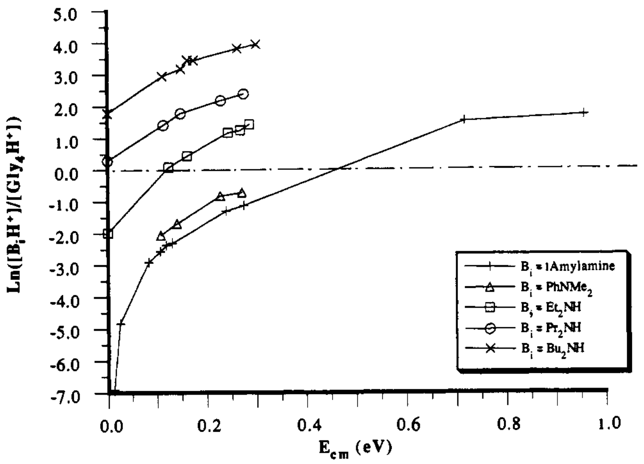

Figure 4. Plot of the logarithms of the ratios of product ions from dissociation of proton-bound dimers containing Gly_4 and monoamines against collision energy in the center-of-mass frame.

observed for dimer systems containing monoamines as one component and 1,4-butanediamine, glycylglycylglycine (Gly_3), or glycylglycylglycylglycine (Gly_4) as the other (Figures 2–4). Instead of converging to zero as seen in Figure 1, the logarithms of product ion ratios increase steadily as the collision energy is increased in somewhat parallel fashion regardless of the values at E_{cm} = 0 eV. These values for some of the dimers are clearly seen to cross the $\ln(B_iH^+]/[BH^+]) = 0$ line at fairly low collision energies, which should not happen according to eq 4. Two conclusions can be drawn from Figures 1–4. First, the term Q_i^*/Q^* in eq 3 describing dissociation of proton-bound dimer systems in Figures 2–4 is not unity. On the basis of collision energy dependencies, the term Q_i^*/Q^* in all the three cases should have similar magnitudes. Second, all the results in each of Figures 2–4 show similar trends. This phenomenon indicates the similarities of the Q_i^*/Q^* term among the proton-bound dimers in each of Figures 2–4 and suggests that, at each E_{cm}, the effective temperature is also similar among the proton-bound dimers in each of Figures 2–4.

Figure 5 shows the collision energy dependence for dissociation of $[Gly_3 + Gly_4]H^+$. Comparison of Figure 5 with the previous figures shows that the behavior of $[Gly_3 + Gly_4]H^+$ is qualitatively similar to that of $[c\text{-}C_6H_{11}NH_2 + Me_2NH]H^+$ in Figure 1 but different from that of systems in Figures 2–4. Thus, the difference in $\Delta S°_{H^+}$ between Gly_3 and Gly_4 should be very small, just as the case in $[c\text{-}C_6H_{11}NH_2 + Me_2NH]H^+$. The 4 kcal/mol difference between PAs of Gly_3 and Gly_4 reported previously[1c] using the standard kinetic method is not, therefore, caused by the difference in their $\Delta S°_{H^+}$ values.

There have been other reports of entropy effects encountered in the application of the kinetic method. In a study of acidity of carboxylic acids, Cooks and co-workers[12a] noted that the proton-

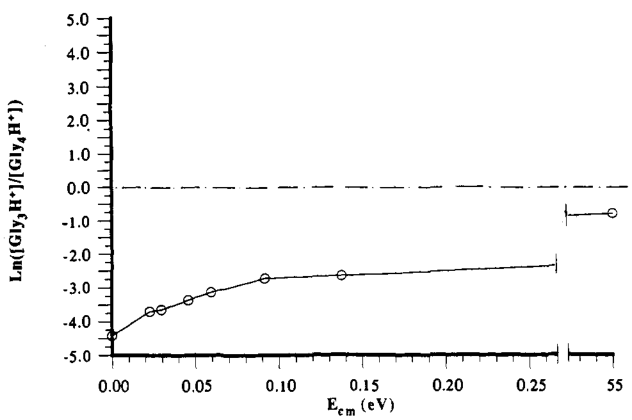

Figure 5. Plot of the logarithms of the ratios of product ions from dissociation of proton-bound dimer $[Gly_3 + Gly_4]H^+$ against collision energy in the center-of-mass frame.

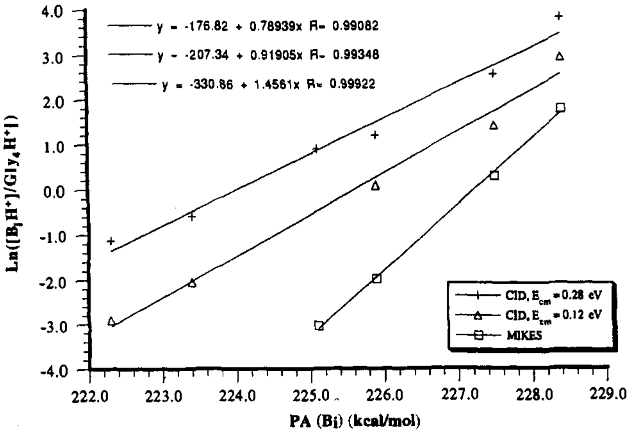

Figure 6. Plot of the logarithms of the ratios of product ions from dissociation of proton-bound dimers containing Gly_4 and monoamines against PAs of the monoamines for experiments done at collision energies 0.28, 0.12, and 0.0 eV in the center-of-mass frame.

bound dimers that contain the formate anion behave differently from other carboxylates studied, and they suggested the involvement of a cyclic structure for the proton-bound dimers that contain formate. In another study using the kinetic method, Cooks and co-workers[12b] observed that pyridine and phenylethylamine were off the line correlating other bases. When the ratio of product ion abundance of pyridine vs *sec*-butylamine was studied at two different collision energies (effective temperature = 381 and 757 K), the changes in the ratio were consistent with the result calculated using the literature value of PA and $\Delta S^\circ_{H^+}$. In a recent publication, Cooks and co-workers[16] examined the dissociation behavior of proton-bound dimers of alcohols with different degrees of substitution. They found that the effective temperature could be very different for different alcohol families and attributed these differences to the entropy effects in different dimer systems. The present work is the first systematic study of the effects of collision energy on the dissociation of proton-bound dimers that include both systems that are expected to show no entropy effects and those that are expected to show strong entropy effects.

When the points in Figure 4 for $E_{cm} = 0$ (MIKE), $E_{cm} = 0.12$, and $E_{cm} = 0.28$ eV were plotted against the PA values of the reference bases, the results fit three straight lines (Figure 6). The quality of the fitting indicates that, at each collision energy (effective temperature), the term Q_i^*/Q^* is roughly the same for each member of B_i and renders quantitative support for the conclusions drawn before by inspection of Figures 1–4. This result is also expected from several reports of successful use of the kinetic method employing CID at different collision energies and/or different pressures of the target gas within each work.[12,17] However, when the attempt was made to extract the PA of Gly_4 using eq 4, different results were obtained for data at different collision energies, as shown in Table II. Specifically, lower apparent PAs were obtained from data at higher collision energies, which indicates that the term Q_i^*/Q^* in eq 3 is larger than unity in this case and that the dissociation of $[B_i + Gly_4]H^+$ to form B_iH^+ is entropically favored over that to form Gly_4H^+.

Table II. Thermochemical Data from Product Ratios in the Dissociation of Proton-Bound Dimers at Various Collision Energies Obtained Using Eq 4

B	E_{cm} (eV)	slope	intercept	T^a (K)	PA^b (kcal/mol)
1,4-butanediamine	0.00	1.719	−400.5	292.7	233.0
	0.10	1.370	−318.0	367.4	232.1
	0.40	1.356	−313.8	371.1	231.4
Gly_3	0.00	1.154	−257.0	436.1	222.7
	0.10	0.915	−203.0	549.8	221.7
	0.30	0.880	−194.0	571.7	220.3
Gly_4	0.00	1.456	−330.9	345.6	227.2
	0.12	0.919	−207.3	547.6	225.6
	0.28	0.789	−176.8	637.5	224.0

[a] Effective temperature of the dimer ion. [b] Apparent proton affinity.

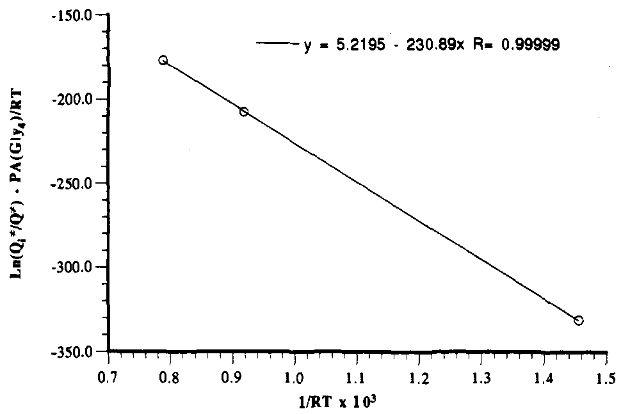

Figure 7. Plot of $[\ln(Q_i^*/Q^*) - PA(B)/RT]$ against $1/RT$ for dissociation of proton-bound dimers containing Gly_4 and monoamines at collision energies 0.28, 0.12, and 0.0 eV in the center-of-mass frame.

Using the assumption that Q_i^*/Q^* is constant within the range of internal energies (effective temperature) of the dimer systems studied, the values of PA and $\ln(Q_i^*/Q^*)$ can be obtained according to eq 5 by plotting $[\ln(Q_i^*/Q^*) - PA(B)/RT_j]$, the intercepts in Figure 6, against $1/RT_j$, the slopes in Figure 6. The final plot is shown in Figure 7. The slope in Figure 7 gives the negative of PA(B) and the intercept gives the value of $\ln(Q_i^*/Q^*)$. With the assumption that there is no reverse barrier for the proton-bound dimer dissociation, the difference in the entropy of protonation between monoamines and Gly_4, $\Delta(\Delta S^\circ_{H^+})$ can be calculated as $\Delta(\Delta S^\circ_{H^+}) = R \ln(Q_i^*/Q^*)$. The numerical results for PA and $\Delta(\Delta S^\circ_{H^+})$ obtained from these analyses are given in Table III for 1,4-butanediamine, Gly_3, and Gly_4. These results again confirm the conclusion drawn before by inspection of Figures 1–4 that the Q_i^*/Q^* terms in eq 3 for systems in Figures 2–4 are different from unity.

As shown in Tables II and III, for all three systems in Figures 2–4, the apparent PAs obtained using eq 4 decrease when the collision energy is increased and all of them are lower than the PA values obtained using eq 5. Another conclusion that can be drawn from these results is that when Q_i^*/Q^* is different from unity, the apparent PA obtained using eq 4 will be closer to the true PA when the experiments are done with lower internal

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Table III. Thermochemical Data from Product Ratios in the Dissociation of Proton-Bound Dimers at Various Collision Energies Obtained Using Eq 5

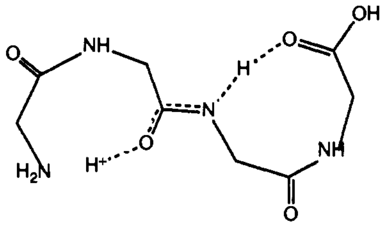
	PA (kcal/mol)		$\Delta(\Delta S^\circ_{H^+})$[a] (eu)	
B	this work	lit.	this work	lit.
1,4-butanediamine	237.5	237.6[b]	15.6	17.1[c]
Gly_3	228.9	223.1[d]	14.2	NA
Gly_4	230.9	227.2[d]	10.4	NA

[a] Difference in entropy of protonation between monoamines and base B. [b] Reference 4; data from equilibrium method. [c] Reference 19c; data from equilibrium method. [d] Reference 1e; data from kinetic method.

energies (effective temperature) of the dimer ions. This can be seen by examining the first two terms in the right-hand side of eq 3, which denote the apparent PA. The importance of the first term, $\ln(Q_i^{\ddagger}/Q^{\ddagger})$, will decrease as the effective temperature decreases. The differences among PAs from the MIKE method using eq 4 and those from eq 5 are 3–5 kcal/mol for the three systems studied in this work (Tables II and III).

The values of PA and $\Delta(\Delta S^\circ_{H^+})$ for 1,4-butanediamine obtained using the procedures developed in this work agree well with the corresponding literature values (Table III). Considering the assumptions made in the kinetic method and, also in the procedures developed in this work, the agreement could be fortuitous. However, since Figures 1 and 2 established unambiguously that 1,4-butanediamine shows a measurable entropy effect in proton-bound dimer dissociation, the agreement may indicate that the assumptions that the $Q_i^{\ddagger}/Q^{\ddagger}$ term is constant in the range of dimer internal energies studied and that there is no reverse barrier are valid in this system. It is noticed that the structures of the proton-bound dimers between bases and peptides are not known and are unlikely to be simple.[18] The binding energy in such complexes can be higher than estimated for simple electrostatic interactions.[18] These two factors will make the assumptions used in the procedures of this work suspicious for dimer systems containing peptides, and the numerical values reported should be regarded as estimates.

Proton affinity increase due to intramolecular H-bonding is a well-known phenomenon. The most familiar examples are probably the α,ω-diamine[19] and polyether[20] systems. A decrease in $\Delta S^\circ_{H^+}$ is another consequence of intramolecular H-bonding. In the case of 1,4-butanediamine, the formation of intramolecular H-bonding is considered to be responsible for an increase of 17.9 kcal/mol in PA and a decrease of 17.1 eu in $\Delta S^\circ_{H^+}$ in comparison with the values for butylamine.[19c] In a system relevant to peptides, CH_3CO-Ala-OEt, an increase of PA by ≈ 7 kcal/mol and a decrease of $\Delta S^\circ_{H^+}$ by ≈ 15 eu relative to simple amides were observed and considered to indicate the occurrence of intramolecular H-bonding.[18a] In a previous paper,[1e] we reported the increase in apparent PA in oligoglycines when the number of the glycine units was increased and suggested intramolecular H-bonding as one of the possible explanations. The present work provides experimental support for the existence of intramolecular H-bonding in Gly_3 and Gly_4. However, the possible structures (conformations) of the protonated peptides are probably large in number. It is possible that in a real situation, these small protonated peptides exist in the gas phase in a number of conformations, each of which fulfills the requirement of stabilizing the proton and the whole system through a network of intramolecular H-bonding and other interactions. One such possible conformation of Gly_4 is shown as follows:



Conclusion

The product ion ratios from dissociation of proton-bound dimers activated at various collision energies were used to examine the effect of entropy on dissociation. By comparison with reference systems of known PA and $\Delta S^\circ_{H^+}$, the peptide systems Gly_3 and Gly_4 were shown to behave more like 1,4-butanediamine, but different from Me_2NH. Using the assumption that the ratio of the partition functions for the activated complexes leading to the two competing dissociation products, $Q_i^{\ddagger}/Q^{\ddagger}$, is constant in the range of the dimer internal energies studied, the values of PA and $R\ \ln(Q_i^{\ddagger}/Q^{\ddagger})$ were obtained by linear regression and were consistent with literature values of PA and $\Delta(\Delta S^\circ_{H^+})$ for 1,4-butanediamine. The values of $R\ \ln(Q_i^{\ddagger}/Q^{\ddagger})$ for Gly_3 and Gly_4 were similar in magnitude to that for 1,4-butanediamine.

The experimental results (collision energy dependence of product ion ratios from dissociation of proton-bound dimers) of this work establish that Gly_3 and Gly_4 show entropy effects in proton-bound dimer dissociation and indicate the existence of intramolecular H-bonding in these protonated peptides in the gas phase. The experimental procedure developed in this work as a semiquantitative way of assessing entropy effects should be applicable to any gas-phase ionic system for which the proton-bound dimer can be generated. Ions from large biomolecules are obvious candidates. The procedure to extract quantitative values for PA and $R\ \ln(Q_i^{\ddagger}/Q^{\ddagger})$ or $\Delta(\Delta S^\circ_{H^+})$ developed in this work, however, involves assumptions in addition to those made in the standard kinetic method, and the numerical results should therefore be presented as estimates of these thermochemical values.

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