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A: Spectroscopy, Molecular Structure, and Quantum Chemistry

**Structure of Proton-Bound Methionine and Tryptophan
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Spectroscopy and Quantum Chemical Calculations**Åke Andersson, Mathias Poline, Meena Kodambattil, Oleksii
Rebrov, Estelle Loire, Philippe Maitre, and Vitali Zhaunerchik*J. Phys. Chem. A*, **Just Accepted Manuscript** • DOI: 10.1021/acs.jpca.9b11811 • Publication Date (Web): 27 Feb 2020Downloaded from pubs.acs.org on February 28, 2020**Just Accepted**

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Structure of Proton-bound Methionine and Tryptophan Dimers in the Gas Phase Investigated with IRMPD Spectroscopy and Quantum Chemical Calculations

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Abstract

The structures of three proton-bound dimers: Met_2H^+ , MetTrpH^+ , and Trp_2H^+ are investigated in the gas phase with infrared multiple photon disassociation (IRMPD) spectroscopy in combination with quantum chemical calculations. Their IRMPD spectra in the range of $600 - 1850 \text{ cm}^{-1}$ are obtained experimentally using the FT-ICR mass spectrometer and the CLIO free electron laser as an IR light source. The most abundant conformers are elucidated by comparing the IRMPD spectra with harmonic frequencies obtained at the B3LYP-GD3BJ/6-311++G** level of theory. Discrepancies between the experimental and theoretical data in the region of $1500 - 1700 \text{ cm}^{-1}$ are attributed to the anharmonicity of the amino bending modes. We confirm the result of a previous IRMPD study that the structure of gas phase Trp_2H^+ is charge-solvated, but find that there are more stable structures than originally reported.¹ In addition, gas phase Met_2H^+ and MetTrpH^+ have been revealed to have charge-solvated structures. For all three dimers, the most stable conformer is found to be of type A. The spectrum of Met_2H^+ , however, cannot be explained without some abundance of type B charge-solvated conformers as well as salt-bridged structures.

Introduction

Amino acids are ubiquitous building blocks for life. They consist of amino group, carboxyl group and a side chain which determines properties of the specific amino acid. In solution, interactions with the solvent molecules cause amino acids to take on a zwitterionic form, meaning that a proton has moved from the carboxyl to the amino functional group. In the context of gas phase studies of isolated and microsolvated amino acids, the question of when and how amino acids become zwitterions has been studied widely.¹⁻²⁹ In many of these studies, infrared multiple photon disassociation (IRMPD) spectroscopy has been used as an experimental tool for molecular structure elucidation.

Protonated alpha amino acid dimers in the gas phase are of interest because they capture

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3 the essence of amino acid interaction while being of relatively small size. They exhibit
4 many conformers, commonly classified as charge-solvated (CS) or salt-bridge (SB) structures.
5 The CS implies binding of the monomers through an inter-molecular hydrogen bond (H-
6 bond) between the protonated amino group of one amino acid and amino group (type A) or
7 carboxyl group (type B) of another amino acid. In the SB structures one monomer adopts
8 a zwitterionic (type Z) form.
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11 The stability of SB structures is related to the stability of monomer's zwitterionic form,
12 which in turn has been attributed to various factors. Wyttenbach *et al.*² found a positive
13 correlation between the stability of the zwitterionic form of alkali metal cationized amino acid
14 monomers and proton affinity (PA) of the amino acid. Bush *et al.*³ found this correlation to
15 be weaker for alkali metal cationized lysine with additional methyl groups, and argued that
16 PA is not a reliable indication of zwitterionic stability whereas side chain effects are more
17 important. By contrast, studies of proton bound amino acid homodimers have shown that
18 zwitterionic stability is well predicted by PA.⁴⁻¹¹ High PA amino acids, *e.g.* arginine, lysine,
19 and proline, have been shown to form SB dimers.⁴⁻⁶ On the other hand, low PA amino acids,
20 *e.g.* glycine, threonine, phenylalanine, and tyrosine, form CS dimers.⁷⁻¹⁰ With a mid PA,
21 glutamic acid has been shown to adopt CS structures at 15 K, but SB dimers at 300 K.¹¹ A
22 recent study by Feng *et al.*¹ of tryptophan dimers has shown it to favor CS over SB, even
23 though its PA is similar to that of proline.
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41 In this paper we report the results of IRMPD studies performed for Met_2H^+ , MetTrpH^+ ,
42 and Trp_2H^+ in the frequency range of 600–1850 cm^{-1} . The tryptophan (Trp, W) homodimer
43 is studied with the aim of revealing its IRMPD spectrum in a previously unexplored frequency
44 range.¹ Studies of the methionine (Met, M) homodimer are motivated by the fact that its PA
45 lies in the frontier of amino acids that are known to form CS structures.^{10,30} An additional
46 reason to study Met is its side chain. Dimers of amino acids tend to stabilize their geometry
47 through intermolecular interaction, *e.g.* H-bonding between side chains, in addition to the
48 CS or SB binding. Methionine has no N, O, or F on its side chain, only S, which forms
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3 significantly weaker H-bonds.^{31,32} It is therefore of interest to study how methionine dimers
4 stabilize. It is also of interest to study Met heterodimers, where the second monomer is prone
5 to intermolecular interactions. For this purpose, the MetTrpH⁺ dimer is studied, which only
6 has the Trp side chain capable of significant intermolecular interaction, presumably N-H···N
7 and N-H··· π to the indole ring.
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Experiment

All experimental data was collected at Centre Laser Infrarouge d'Orsay (CLIO) in Orsay, France. Three amino acid dimers were studied: Met₂H⁺, MetTrpH⁺, and Trp₂H⁺. The Trp used for Trp₂H⁺ was an isotopologue, specifically five deuterium atoms were substituted on the indole ring. The dimer solutions were prepared as 1 mM monomer concentration in a 49:49:2 mixture of water, methanol, and formic acid. Ions were delivered into the gas phase by electrospray ionization (ESI) of the solution, and subsequently stored and accumulated in a linear ion-trap of a mass spectrometer (MS) of model 7 T Bruker Apex Qe. While trapped, ions were subsequently pulse extracted toward the ion cyclotron resonance (ICR) ion trap, where they were irradiated with pulses of the CLIO free-electron laser (FEL), which scanned the 600 – 1850 cm⁻¹ frequency range. The FEL had power 20 – 30 mW and pulse rate 25 Hz. Irradiation time of the ions was controlled by a mechanical shutter placed between the FEL beam and the ICR ion trap. It is typically chosen to be around 500 ms. The measurements were performed at high, mid, and low irradiation energy, meaning the summed energy of the FEL pulses over the irradiation time was above, in, and below the range of 8 – 16 mJ, respectively. Finally, the abundance of dimers and their fragments were analyzed using Fourier transform ICR in the MS.

For a given FEL frequency, the IRMPD intensity was calculated as:

$$I_{\text{IRMPD}} = \ln\left(1 + \frac{I_{\text{fragment}}}{I_{\text{parent}}}\right),$$

where $I_{\langle\text{species}\rangle}$ is the integrated MS intensity of that species and its isotopologues. The three dimers Met_2H^+ , MetTrpH^+ , and Trp_2H^+ respectively fragment into MetH^+ , TrpH^+ , and TrpH^+ . With high irradiation, these may fragment further. In that case, I_{fragment} is replaced by the sum of all fragments' intensities.

Calculations

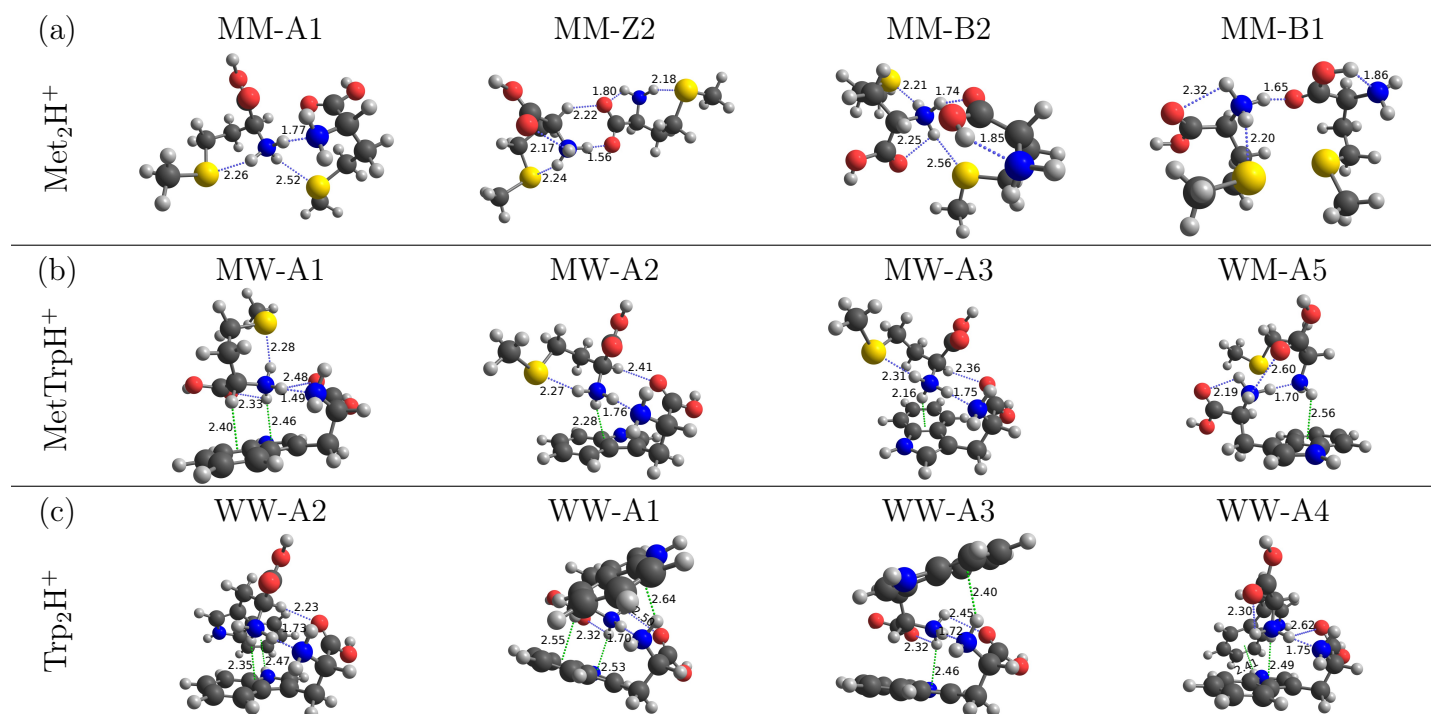


Figure 1: The conformers of (a) Met_2H^+ , (b) MetTrpH^+ , and (c) Trp_2H^+ with the lowest Gibbs energies at $T = 300$ K. The distances in Å of possible H-bonds (blue dotted lines) and cation- π interactions (dotted green lines) are shown. The strength of these interactions are investigated in our NCI analysis (fig. S4, S5, and S6 in SI). The shorthand XY-SN denotes a dimer structure. X and Y are one-letter amino acid codes of the protonated and unprotonated moiety, respectively. S is the structure type, which can be A, B, or Z. N is the index when sorted among structures of the same type, with respect to electronic energy at the CBS-4M level of theory.

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3 A conformational search has been performed using several molecular dynamics (MD)
4 simulations with different initial geometries in order to cover a large conformation space.
5 The MD calculations were carried out in the microcanonical ensemble employing the density
6 functional-based tight binding method³³ as implemented in the DFTB+ software package.³⁴
7 The initial velocities were considered to correspond to a Maxwell-Boltzmann distribution at
8 298 K and a velocity Verlet algorithm with a time step of 1 fs was implemented. For the
9 amino acid heterodimer, MetTrpH⁺, we considered two sets of structures with either Met or
10 Trp moiety being protonated.
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13 The most stable structures obtained with the MD simulations were optimized with DFT
14 using the B3LYP functional and 6-311++G** basis set including the GD3BJ empirical dis-
15 persion. The harmonic frequency analysis was performed at the same level of theory. For
16 the optimized structures single point energies were calculated with the CBS-4M composite
17 method. To obtain the corresponding Gibbs energies, the CBS-4M calculations were com-
18 bined with vibrational analysis performed with the B3LYP-GD3BJ/6-311++G** method.
19 For Met₂H⁺ dimers, we additionally performed VPT2 anharmonic vibrational analysis as well
20 as single point energy calculations at the G4MP2 level of theory. To reduce computational
21 costs, the VPT2 analysis was performed with the N07D basis set, which has been shown
22 to perform well under similar circumstances.³⁵ We note that for MetTrpH⁺ and Trp₂H⁺
23 dimers, which have more complex structures, neither G4MP2 calculations nor the VPT2
24 analysis were undertaken due to limitations in computational time. All these calculations
25 were carried out with the Gaussian 16 program.³⁶
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45 When comparing the harmonic frequency analysis of conformers with an experimental
46 spectrum, three modifications are made. First, all frequencies are multiplied by 0.980, to
47 account for anharmonicity. This frequency scaling constant was found by least-square fitting
48 harmonic theory to all experimental spectra, and is consistent with B3LYP frequency scaling
49 for other molecules.³⁷ Second, for every conformer, its vibrational frequencies are broadened
50 into a continuous spectrum by convolution with a Gaussian function with parameter $\sigma =$
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8 cm⁻¹, or equivalently with a full width at half maximum of 18.8 cm⁻¹. This broadening was required in order to take into account the FEL pulse linewidth. Last, the spectra of conformers are scaled in proportion to their relative abundances at room temperature, and summed. The relative abundances p_i were found as

$$p_i = \frac{1}{Z} \exp\left(-\frac{G_i}{k_B T}\right); \quad Z = \sum_i \exp\left(-\frac{G_i}{k_B T}\right), \quad (1)$$

where G_i are the Gibbs energies of the conformers, $k_B = 0.6950$ cm⁻¹/K is the Boltzmann constant, and T is the temperature.

With the goal of understanding and classifying the inter- and weak intramolecular interactions, noncovalent interaction (NCI) analyses were performed for the most abundant conformers of Met₂H⁺, MetTrpH⁺, and Trp₂H⁺. The NCI method introduced by Johnson *et al.*³⁸ uses the electron density and its gradient to classify real-space regions as van der Waals interactions, H-bonds, or steric repulsions. The $\text{sign}(\lambda_2)\rho$ index assigned by the NCI method, where ρ is the electron density and λ_2 is the second largest eigenvalue of its Hessian, can also be related to the binding energy of H-bonds.³⁹

Results and discussion

Methionine-methionine dimers

The four most abundant structures of Met₂H⁺ at $T = 300$ K are shown in fig. 1a. As inferred from our NCI analyses (fig. S4 in SI), in all four, the S on the protonated moiety forms an intramolecular H-bond with the protonated amino group, causing the side chain to bend. In MM-A1 and MM-B2, the other S weakly interacts with the protonated amino group, stabilizing the dimer further. In MM-B1, MM-B2, and MM-Z2, an H-bond is formed between the amino and carboxyl group of the unprotonated moiety.

The single point energies of Met₂H⁺ were calculated at three levels of theory. In order of

increasing accuracy and computational expense, they are: B3LYP, CBS-4M, and G4MP2. An energy comparison of the methods is made in fig. 2. Compared to the composite methods

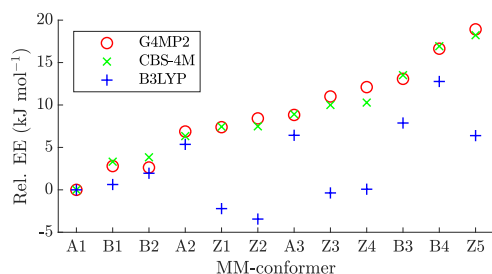


Figure 2: Electronic energy for Met_2H^+ conformers relative to MM-A1, calculated at three levels of theory. The geometries of all Met_2H^+ conformers are available as Supporting Information (fig. S1 in SI).

(CBS-4M and G4MP2), B3LYP greatly underestimates the energy of SB conformers. A similar tendency was previously reported in studies of serine dimers.²⁰ The CBS-4M electronic energy calculations of Met_2H^+ conformers give results consistent with G4MP2, while having the advantage of being less time consuming. In light of this agreement, CBS-4M is assumed to be suitable for energy calculations for both other proton-bound dimers studied in this paper. The Gibbs energies of Met_2H^+ conformers were therefore calculated with CBS-4M, and the corresponding relative abundances are shown in fig. 3a.

The IRMPD spectra of Met_2H^+ with different irradiation energies are shown in fig. 4, together with harmonic frequency analyses for the four most abundant conformers. The Gibbs energy analysis based on eq. (1) predicts the MM-A1 conformer to be the most abundant species, though its relative population constitutes less than 40%. This is consistent with the experimental data, *i.e.* by comparing the IRMPD spectrum near 1405 cm^{-1} to the harmonic frequency analyses, MM-A1 can be ruled out to be the only populated conformer. Predictions for MM-Z2 lack the second peak observed in the region of $1700 - 1800\text{ cm}^{-1}$ associated with carboxyl group stretching, which is explained by the fact that the peaks in this region correspond to vibrational modes in the carboxyl group, whereas the type Z conformer possesses only one carboxyl group. The observed peak at 1725 cm^{-1} being unmatched by MM-A1 and MM-Z2, leads us to believe that there is a substantial abundance of MM-B2

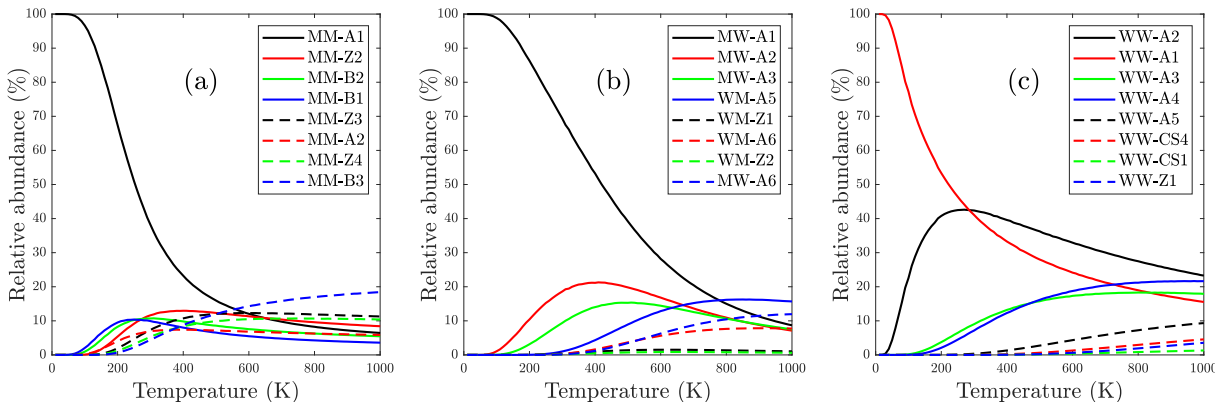


Figure 3: Temperature-dependent relative abundances for the eight most abundant (at 300 K) conformers of (a) Met₂H⁺, (b) MetTrpH⁺, and (c) Trp₂H⁺, obtained with eq. (1) employing the CBS-4M method for calculating Gibbs energies. The relative abundances do not sum to exactly one because additional conformers were considered when calculating the thermodynamic partition function. Note that because conformers are indexed by type and electronic energies, not by type and Gibbs energies, some low-index conformers *e.g.* WM-A1 are not among the eight most abundant conformers.

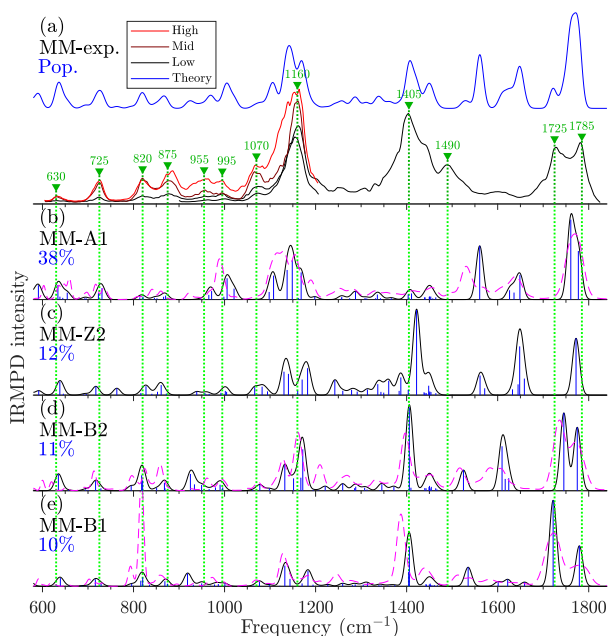


Figure 4: IRMPD spectra of Met₂H⁺. (a) The experimental IRMPD spectra measured at different irradiation energies (black/wine/red solid lines), with local maxima highlighted (green triangles) and traced. The sum of predicted spectra of conformers weighted by relative abundances from fig. 3a (blue line) is included for comparison, offset to ease viewing. (b-e) Scaled harmonic frequencies (solid blue lines) of individual conformers and their relative abundance (blue text) obtained from eq. (1). Predicted spectra (black solid lines) were obtained by convoluting with a Gaussian function with FWHM = 18.8 cm⁻¹. The anharmonic VPT2 alternative (dashed magenta lines) is included for comparison.

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3 or MM-B1, which both agree well with the experimental IRMPD spectrum. This is again
4 consistent with the Gibbs energy population analysis, which predicts their relative popula-
5 tions to be 11% and 10%, respectively. All conformers except for MM-B1 are predicted to
6 have large peaks in the region of $1500 - 1700 \text{ cm}^{-1}$, but the IRMPD intensity in this region
7 is contrastingly low and flat. The frequencies in this region correspond to bending modes
8 of the amino group. Because the amino group participates in the H-bonding of the dimer
9 and the potential along the proton transfer coordinate is quite flat, we believe these modes
10 to be strongly anharmonic, and harmonic analyses to be insufficient. This anharmonicity
11 is known to play a significant role for H-bonding amino stretching in Lys_2H^+ ,^{40,41} so it is
12 plausible that bending also is affected.

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15 In order to shed more light on the discrepancies in the region of $1500 - 1700 \text{ cm}^{-1}$, a
16 VPT2 anharmonic vibrational analysis was performed. The resulting spectra are shown as
17 dashed magenta lines in fig. 4. The anharmonic analysis of MM-Z2 did not produce a sensible
18 result. For abundant conformers of type A and B, the anharmonic analyses indeed predict
19 flatter and less intense IRMPD spectra in the region of $1500 - 1700 \text{ cm}^{-1}$. Furthermore, the
20 VPT2 bands are red-shifted compared to those predicted by the harmonic analysis. Overall,
21 the anharmonic treatment improves agreement with the experiment.

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24 It is interesting to note that the VPT2 bands of MM-B2 fit the experimental spectra
25 better than those of MM-A1, calling into the question of whether MM-A1 truly is the most
26 abundant. This can point to the non-thermal population of conformers produced upon
27 ESI. Although this phenomenon was observed in previous studies performed with other
28 experimental setups,⁴² this scenario is very unlikely in our case. When the ions accumulate
29 in the linear ion trap of the ICR, multiple low energy collisions with the Ar buffer gas
30 atoms occur which leads to a thermalization of the ions. On the other hand, a relatively
31 low predicted abundance for MM-B2 conformer might point to deficiencies of the theoretical
32 calculations.

Methionine-tryptophan dimers

The four most abundant structures of MetTrpH^+ at $T = 300\text{ K}$ are shown in fig. 1b. All structures shown are of type A, and, according to our NCI analyses (fig. S5 in SI), are additionally stabilized by a cation- π interaction between the amino group of the Met moiety and the indole ring. A weak intermolecular interaction exists between either the amino group (MW-A1, WM-A5) or the α -H (MW-A2, MW-A3) of the protonated moiety and carboxyl group. For the case when the Met moiety is protonated, its side chain bends due to an H-bond between the S and the amino group, similar to the Met_2H^+ case. In MW-A1, there is an additional cation- π interaction between Met α -H and the phenyl part of the indole ring, making MW-A1 the most stable structure by far.

Following the conclusion drawn from the Met_2H^+ results, the electronic energies of MetTrpH^+ were calculated with CBS-4M, and are shown in tab. 1. The Gibbs energies

Table 1: Relative electronic energies (EE) of the most stable MetTrpH^+ and labeled Trp_2H^+ conformers, calculated with CBS-4M. The first letter in dimer notation is the name of the amino acid where the proton is. The geometries of all MetTrpH^+ and Trp_2H^+ conformers are available as Supporting Information (fig. S2 and S3 in SI, respectively).

Conf.	EE (kJ mol ⁻¹)	Conf.	EE (kJ mol ⁻¹)
MW-A1	0.00	WW-A1	0.00
MW-A2	4.67	WW-A2	1.14
MW-A3	7.06	WW-A3	5.87
WM-A1	10.55	WW-A4	6.89
WM-Z1	14.98	WW-A5	15.10
WM-A2	15.76	WW-CS4 ¹	21.26
WM-A3	15.77	WW-CS1 ¹	21.49
WM-Z2	15.93	WW-A6	29.68
WM-A4	16.44	WW-Z1	31.33
WM-A5	17.41	WW-B1	40.43

of MetTrpH^+ conformers were calculated, and the corresponding relative abundances are shown in fig. 3b. As it follows from the figure, the MW-A1 conformer is the most abundant species in a broad temperature range.

The IRMPD spectra of MetTrpH^+ for different irradiation energies are shown in fig. 5,

together with harmonic frequency analyses for the four most abundant conformers and the most abundant conformers of type B and Z. The experimental IRMPD spectra show that

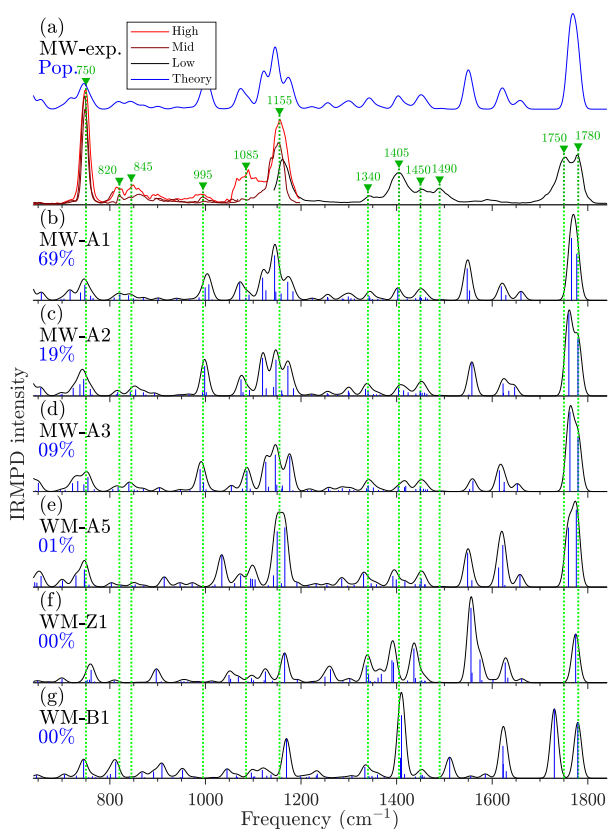


Figure 5: Same as fig. 4, but for MetTrpH⁺ and without VPT2. WM-Z1 and WM-B1 are ranked 5 and 16 in abundance, respectively.

there is a large peak at 750 cm⁻¹, substantially larger than the one at 995 cm⁻¹. According to the harmonic frequency analysis, all conformers are predicted to have a peak near 750 cm⁻¹, but only the three most abundant conformers are predicted to have a peak near 995 cm⁻¹, though of magnitude similar to the 750 cm⁻¹ peak. The experimental scan with high irradiation energy revealed a peak at 1085 cm⁻¹, which is only present for the conformers of type A. As was the case for Met₂H⁺, the type Z conformer is predicted to have only a single peak in the region of 1700 – 1800 cm⁻¹. Therefore, it is unlikely to be significantly populated, which is in line with the population analysis based on eq. (1). Again, we observe low and flat IRMPD intensity in the region of 1500 – 1700 cm⁻¹, but harmonic analyses predict relatively intense peaks there. Anharmonic analyses were not undertaken due to limitations on com-

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putational time, but we hypothesize that the peaks in the region of $1500 - 1700 \text{ cm}^{-1}$ would have become less intense, as they did for Met_2H^+ . The harmonic analyses for the three most abundant conformers all fit the experimental spectra adequately, which is consistent with the populations obtained from the Gibbs energies using eq. (1).

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It is interesting to note that despite the fact that in the most abundant MetTrpH^+ conformers the proton is located on the Met moiety, as seen in fig. 1b, the IRMPD mass spectra of MetTrpH^+ do not show MetH^+ fragments, only TrpH^+ . A plausible explanation can be suggested based on the multi-photon nature of the IRMPD process. The successive photon absorption is not instantaneous, since it is determined by the intramolecular vibrational energy redistribution time scale. This implies that the dimer can undergo conformational conversion such that the proton might migrate from Met to the Trp moiety. Another explanation for this observation could be that the $\text{TrpH}^+ + \text{Met}$ fragmentation channel is more energetically favorable than $\text{MetH}^+ + \text{Trp}$. In order to justify this explanation, we calculated energies of these channels as difference between enthalpies at $T = 300 \text{ K}$ of the corresponding fragments and the parent dimer assuming the lowest energy conformer, MW-A1. The enthalpies for MetH^+ , TrpH^+ , Met and Trp were found with the CBS-4M method (single point energy calculation) for which the geometries were optimized at the B3LYP-GD3BJ/6-311++G** level of theory. According to our calculations the energies of the $\text{TrpH}^+ + \text{Met}$ and $\text{MetH}^+ + \text{Trp}$ channels are 158 and 160 kJ mol^{-1} , respectively. Although the $\text{TrpH}^+ + \text{Met}$ channel is indeed energetically more favorable, the difference of only 200 J mol^{-1} is not likely to explain the predominance of the $\text{TrpH}^+ + \text{Met}$ fragmentation channel. Therefore we believe that the absence of the MetH^+ fragment in the mass spectra provides evidence for isomerization occurring during the IRMPD process. Isomerization has been observed in the IRMPD process for other ions,^{43,44} and thus it is not so surprising that it also occurs for proton-bound dimers, which have almost no barrier for the proton transfer.⁴⁵

Tryptophan-tryptophan dimers

The four most stable structures of Trp_2H^+ at $T = 300\text{ K}$ are shown in fig. 1c. They are all of type A, and are additionally stabilized by cation- π as well as intermolecular H interaction, as is inferred from the NCI analyses (fig. S6 in SI). In addition to the characteristic H-bond of the type A structures, every structure has a stabilizing weak interaction, between either the amino group (WW-A1, WW-A3, WW-A4) or the α -H (WW-A2) of the protonated moiety and carboxyl group. WW-A1 is set apart from the rest by having three relatively strong stabilizing cation- π interactions from each amino group to the indole N on the other moiety.

The electronic energies of Trp_2H^+ were calculated with CBS-4M, and are listed in tab. 1. Two structures previously reported,¹ WW-CS1 and WW-CS4, are included among those found by our conformational search. The Gibbs energies of Trp_2H^+ conformers were calculated, and the corresponding relative abundances are shown in fig. 3c. It is interesting to note that even though WW-A1 is the most stable at low temperatures, WW-A2 becomes slightly more abundant at $T = 300\text{ K}$. Such a behavior can be explained in terms of entropy. The many cation- π interaction in WW-A1 constrain the side chains and limits the entropy, while the relative floppiness of WW-A2 allows for a higher entropy. Our calculations show that the entropy of WW-A1 is indeed $0.6k_B$ ($5\text{ J mol}^{-1}\text{ K}^{-1}$) lower than that of WW-A2.

For Trp_2H^+ the seven most abundant conformers are of type A. Feng *et al.*¹ have already asserted that the most abundant conformers of Trp_2H^+ are of type A. The most stable conformer found in the current paper has electronic energy 21.26 kJ mol^{-1} and Gibbs energy 17.96 kJ mol^{-1} less than WW-CS4, which we find to be the most stable conformer among those previously reported. To rule out the possibility that the discrepancy between the findings of Feng *et al.* and ours is caused by using different numerical methods, we also calculated electronic energies at the same level of theory. Employing the M062X functional as used in Ref.,¹ the electronic and Gibbs energy of WW-A1 are still lower, by 22.97 and 20.29 kJ mol^{-1} , respectively, than that of WW-CS1.

The IRMPD spectra of Trp_2H^+ in the region of $950 - 1850\text{ cm}^{-1}$ for different irradiation

energies are shown in fig. 6, together with harmonic frequency analyses for the four most abundant conformers. Due to issues arising from a contaminant in the MS, the IRMPD spectra below 950 cm^{-1} could not be obtained. Similar to the Met_2H^+ and MetTrpH^+ cases,

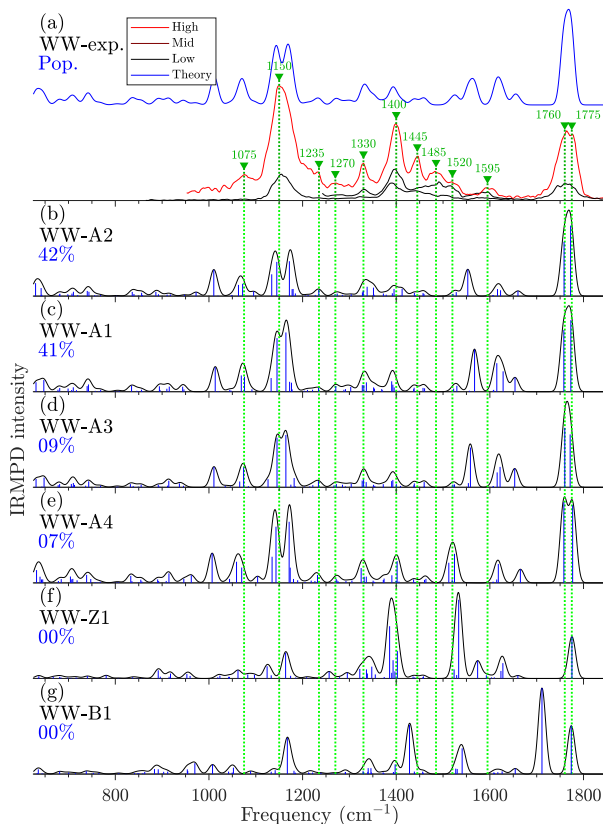


Figure 6: Same as fig. 4, but for Trp_2H^+ and without VPT2. WW-Z1 and WW-B1 are ranked 8 and 9 in abundance, respectively.

there is a reasonable agreement between observations and theoretical predictions outside the region of $1500 - 1700\text{ cm}^{-1}$ for conformers of type A. WW-B1 can be ruled out because of its predicted peak near 1700 cm^{-1} not seen in experimental IRMPD spectra. WW-Z1 has only one peak in the carboxyl region, *i.e.* $1700 - 1800\text{ cm}^{-1}$, while two peaks are actually observed. We thus conclude that the structure of Trp_2H^+ is of type A in the gas phase. Because the most stable structures are all of type A, their predicted IRMPD spectra are quite similar (fig. 6b-e), and their relative abundances cannot be inferred from the experimental IRMPD spectra unambiguously. Nonetheless, employing abundances based on the Gibbs energy analysis eq. (1), the combined theoretical spectrum (blue line in fig. 6a) agrees reasonably

well with the experimental IRMPD spectrum.

Conclusions

In this paper, we investigated the structure of Met_2H^+ , MetTrpH^+ , and Trp_2H^+ dimers. For this purpose, we employed IRMPD spectroscopy in combination with theoretical calculations. Based on the comparison of the observed IRMPD spectra with harmonic frequencies of low-energy conformers, scaled by 0.980 to compensate for anharmonicity, it is concluded for all three dimers that the most abundant conformer is CS. For MetTrpH^+ and Trp_2H^+ , we further conclude that the most abundant conformers are of type A, but cannot differentiate the most stable type A conformers, because their predicted spectra were quite similar. In absence of distinct conformer-specific spectral features, the relative abundances of conformers calculated with eq. (1) were used, where Gibbs energies are input parameters. The conformer abundances obtained in such a way provide reasonable agreement of the experimental IRMPD spectra with the combined theoretical IR spectra.

For calculating Gibbs energy, three methods were used for the B3LYP-GD3BJ/6-311++G** optimized geometries: B3LYP, CBS-4M, and G4MP2. It was found that CBS-4M behaved similarly to G4MP2 for Met_2H^+ while being significantly cheaper, and was therefore utilized for all energy calculations. It was also found that B3LYP underestimates energies of the type Z conformers.

Trp_2H^+ was studied previously by Feng *et al.*¹ Our results for Trp_2H^+ confirm that the most stable structure of Trp_2H^+ is of type A, but include five more stable structures, of which the most stable has electronic energy $21.26 \text{ kJ mol}^{-1}$ and Gibbs energy $17.96 \text{ kJ mol}^{-1}$ less than the most stable among those previously reported.

The harmonic frequency analyses adequately describe most of the observed IRMPD features with the exception of those in the region of $1500 - 1700 \text{ cm}^{-1}$, where the experimental IRMPD features are rather weak and flat. The harmonic analyses assign these peaks to amino

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3 bending vibrational modes, and overestimate their intensities. We believe these modes to be
4 anharmonic as they involve hydrogen bonds between amino groups and the potential for the
5 proton hopping between the two moieties is rather flat. Anharmonic analyses using VPT2
6 improves the agreement with the experimental data in the region of $1500 - 1700 \text{ cm}^{-1}$, with
7 the notable exception of MM-Z2, for which VPT2 does not produce a sensible result.
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13 As mentioned in the introduction, the aim of this study was to understand how the side
14 chains stabilize. Cation- π interaction is seen for Trp_2H^+ between the indole ring and any
15 sufficiently positive hydrogen, *e.g.* those adjacent to N and α -H, though the latter is rather
16 uncommon. $\text{S} \cdots \text{H}$ bonding in Met_2H^+ is concluded to occur between S and protonated
17 amino groups. For the heterodimer MetTrpH^+ , side chain stabilization occurs through both.
18 If the Met moiety amino group is protonated, it interacts with (in order of decreasing NCI
19 index ρ) the Trp amino group, S, and indole ring. Otherwise, in many structures including
20 the most abundant, the protonated Trp amino group only interacts with the Met amino and
21 carboxyl group, and the S does not participate in a significant interaction.
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31 Tryptophan and proline may be a remarkable counterexample to the empiric rule which
32 states that the stability of SB structures of homodimers increases with the PA of the
33 monomer.⁸ The uncertainty lies in which of those amino acids have the greater PA, be-
34 cause they are not consistently ranked across works (see Ref.³⁰ and references therein). The
35 same can be said for methionine and proline. We speculate that while the PA of an amino
36 acid is a mostly reliable indicator for whether the homodimer structure is SB, other proper-
37 ties, *e.g.* side chains, affect the structure type when the PA is “on the fence”. This seems to
38 be the case for amino acids which lie near proline on the PA scale.¹⁰
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47 In the future we plan to investigate diastereomer-specific IR features of homo- and het-
48 erochiral amino acid dimers. Such studies are essential for developing analytical tools for
49 characterizing sample enantiomeric purity. The very recent studies on proton-bound glu-
50 tamic acid dimers have demonstrated the experimental evidence for chiral recognition of
51 amino acid dimers from their IR spectra.¹¹ Furthermore, we have recently reported similar
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3 studies on different amino acid dimers from theoretical perspectives which has also veri-
4 fied the potential of this approach.⁴⁶ The results presented in this paper were obtained for
5 homochiral dimers and thus they can readily complement our envisioned studies.
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29 Supporting Information Available

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31 The following files are available free of charge.

- 32 • `supp_info.pdf` : Supporting Information containing figures S1-S6 referred to in this
33 article, and tables of the Cartesian coordinates of each of the 44 conformers.
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43

44 References

- 45 (1) Feng, R.; Yin, H.; Kong, X. Structure of Protonated Tryptophan Dimer in the Gas
46 Phase Investigated by IRPD Spectroscopy and Theoretical Calculations. *Rapid Com-*
47 *mun. Mass Spectrom.* **2016**, *30*, 24–28.
48
49
50
51
52
- 53 (2) Wytttenbach, T.; Witt, M.; Bowers, M. T. On the Stability of Amino Acid Zwitterions
54
55
56
57
58
59
60

- 1
2
3 in the Gas Phase: The Influence of Derivatization, Proton Affinity, and Alkali Ion
4 Addition. *Journal of the American Chemical Society* **2000**, *122*, 3458–3464.
5
6
7
- 8 (3) Bush, M. F.; Oomens, J.; Williams, E. R. Proton Affinity and Zwitterion Stability: New
9 Results from Infrared Spectroscopy and Theory of Cationized Lysine and Analogues
10 in the Gas Phase. *The Journal of Physical Chemistry A* **2009**, *113*, 431–438, PMID:
11 19128186.
12
13
14
15
- 16 (4) Price, W. D.; Jockusch, R. A.; Williams, E. R. Is Arginine a Zwitterion in the Gas
17 Phase? *Journal of the American Chemical Society* **1997**, *119*.
18
19
20
21
- 22 (5) Kong, X. Reinvestigation of the Structure of Protonated Lysine Dimer. *Journal of The*
23 *American Society for Mass Spectrometry* **2014**, *25*, 422–426.
24
25
- 26 (6) Alahmadi, Y. J.; Gholami, A.; Fridgen, T. D. The Protonated and Sodiated dimers of
27 Proline Studied by IRMPD Spectroscopy in the N–H and O–H Stretching Region and
28 Computational Methods. *Phys. Chem. Chem. Phys.* **2014**, *16*, 26855–26863.
29
30
31
32
- 33 (7) Wu, R.; McMahon, T. B. Infrared Multiple Photon Dissociation Spectra of Proline and
34 Glycine Proton-Bound Homodimers. Evidence for Zwitterionic Structure. *Journal of*
35 *the American Chemical Society* **2007**, *129*, 4864–4865, PMID: 17394317.
36
37
38
39
- 40 (8) Yin, H.; Kong, X. Structure of Protonated Threonine Dimers in the Gas Phase: Salt-
41 Bridged or Charge-Solvated? *Journal of The American Society for Mass Spectrometry*
42 **2015**, *26*, 1455–1461.
43
44
45
46
- 47 (9) Kong, X.; Tsai, I.-A.; Sabu, S.; Han, C.-C.; Lee, Y. T.; Chang, H.-C.; Tu, S.-
48 Y.; Kung, A. H.; Wu, C.-C. Progressive Stabilization of Zwitterionic Structures in
49 [H(Ser)_{2–8}]⁺ Studied by Infrared Photodissociation Spectroscopy. *Angewandte Chemie*
50 **2006**, *118*, 4236–4240.
51
52
53
54
55
56
57
58
59
60

- 1
2
3
4
5
6
7
8
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41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60
- (10) Seo, J.; Hoffmann, W.; Malerz, S.; Warnke, S.; Bowers, M. T.; Pagel, K.; von Helden, G. Side-chain Effects on the Structures of Protonated Amino Acid Dimers: A Gas-phase Infrared Spectroscopy Study. *International Journal of Mass Spectrometry* **2018**, *429*, 115 – 120, Terry B. McMahon 70th Birthday Special Issue: Mass Spectrometry and its Application to the Physical Chemistry of Gaseous Ions.
- (11) Klyne, J.; Bouchet, A.; Ishiuchi, S.-i.; Fujii, M.; Schneider, M.; Baldauf, C.; Dopfer, O. Probing Chirality Recognition of Protonated Glutamic Acid Dimers by Gas-phase Vibrational Spectroscopy and First-principles Simulations. *Physical Chemistry Chemical Physics* **2018**, *20*, 28452–28464.
- (12) Price, W. D.; Schnier, P. D.; Williams, E. R. Binding Energies of the Proton-Bound Amino Acid Dimers Gly \oplus Gly, Ala \oplus Ala, Gly \oplus Ala, and Lys \oplus Lys Measured by Black-body Infrared Radiative Dissociation. *The Journal of Physical Chemistry B* **1997**, *101*, 664–673.
- (13) Jockusch, R. A.; Lemoff, A. S.; Williams, E. R. Hydration of Valine- Cation Complexes in the Gas Phase: On the Number of Water Molecules Necessary to Form a Zwitterion. *The Journal of Physical Chemistry A* **2001**, *105*, 10929–10942.
- (14) Lemoff, A. S.; Bush, M. F.; Williams, E. R. Structures of Cationized Proline Analogues: Evidence for the Zwitterionic Form. *The Journal of Physical Chemistry A* **2005**, *109*, 1903–1910.
- (15) Xu, S.; Nilles, J. M.; Bowen Jr, K. H. Zwitterion Formation in Hydrated Amino Acid, Dipole Bound Anions: How Many Water Molecules are Required? *The Journal of chemical physics* **2003**, *119*, 10696–10701.
- (16) Oh, H.; Breuker, K.; Sze, S. K.; Ge, Y.; Carpenter, B. K.; McLafferty, F. W. Secondary and Tertiary Structures of Gaseous Protein Ions Characterized by Electron Eapture

- 1
2
3 Dissociation Mass Spectrometry and Photofragment Spectroscopy. *Proceedings of the*
4 *National Academy of Sciences* **2002**, *99*, 15863–15868.
5
6
7
8 (17) Oh, H.-B.; Lin, C.; Hwang, H. Y.; Zhai, H.; Breuker, K.; Zabrouskov, V.; Carpen-
9 ter, B. K.; McLafferty, F. W. Infrared Photodissociation Spectroscopy of Electrosprayed
10 Ions in a Fourier Transform Mass Spectrometer. *Journal of the American Chemical So-*
11 *ciety* **2005**, *127*, 4076–4083.
12
13
14
15
16 (18) Rodgers, M.; Armentrout, P.; Oomens, J.; Steill, J. Infrared Multiphoton Dissociation
17 Spectroscopy of Cationized Threonine: Effects of Alkali-metal Cation Size on Gas-phase
18 Conformation. *The Journal of Physical Chemistry A* **2008**, *112*, 2258–2267.
19
20
21
22
23 (19) Dunbar, R. C.; Steill, J. D.; Oomens, J. Cationized Phenylalanine Conformations Char-
24 acterized by IRMPD and Computation for Singly and Doubly Charged Ions. *Physical*
25 *Chemistry Chemical Physics* **2010**, *12*, 13383–13393.
26
27
28
29
30 (20) Kong, X.; Tsai, I.-A.; Sabu, S.; Han, C.-C.; Lee, Y. T.; Chang, H.-C.; Tu, S.-Y.;
31 Kung, A.; Wu, C.-C. Progressive Stabilization of Zwitterionic Structures in [H (Ser)
32 2–8]⁺ Studied by Infrared Photodissociation Spectroscopy. *Angewandte Chemie Inter-*
33 *national Edition* **2006**, *45*, 4130–4134.
34
35
36
37
38
39 (21) Kong, X.; Lin, C.; Infusini, G.; Oh, H.-B.; Jiang, H.; Breuker, K.; Wu, C.-C.;
40 Charkin, O. P.; Chang, H.-C.; McLafferty, F. W. Numerous Isomers of Serine Oc-
41 tamer Ions Characterized by Infrared Photodissociation Spectroscopy. *ChemPhysChem*
42 **2009**, *10*, 2603–2606.
43
44
45
46
47
48 (22) Liao, G.; Yang, Y.; Kong, X. Chirality Effects on Proline-substituted Serine Octamers
49 Revealed by Infrared Photodissociation Spectroscopy. *Physical Chemistry Chemical*
50 *Physics* **2014**, *16*, 1554–1558.
51
52
53
54
55 (23) Wu, R.; McMahon, T. B. Stabilization of the Zwitterionic Structure of Proline by
56
57
58
59
60

- 1
2
3 an Alkylammonium Ion in the Gas Phase. *Angewandte Chemie International Edition*
4 **2007**, *46*, 3668–3671.
5
6
7
- 8 (24) Wu, R.; Marta, R. A.; Martens, J. K.; Eldridge, K. R.; McMahon, T. B. Experimental
9 and Theoretical Investigation of the Proton-bound Dimer of Lysine. *Journal of the*
10 *American Society for Mass Spectrometry* **2011**, *22*, 1651.
11
12
13
- 14 (25) Bush, M. F.; Oomens, J.; Saykally, R. J.; Williams, E. R. Effects of Alkaline Earth
15 Metal Ion Complexation on Amino Acid Zwitterion Stability: Results from Infrared
16 Action Spectroscopy. *Journal of the American Chemical Society* **2008**, *130*, 6463–6471.
17
18
19
- 20 (26) Bush, M. F.; Oomens, J.; Williams, E. R. Proton Affinity and Zwitterion Stability: New
21 Results from Infrared Spectroscopy and Theory of Cationized Lysine and Analogues in
22 the Gas Phase. *The Journal of Physical Chemistry A* **2008**, *113*, 431–438.
23
24
25
- 26 (27) O'Brien, J. T.; Prell, J. S.; Steill, J. D.; Oomens, J.; Williams, E. R. Interactions
27 of Mono- and Divalent Metal Ions with Aspartic and Glutamic Acid Investigated with
28 IR Photodissociation Spectroscopy and Theory. *The Journal of Physical Chemistry A*
29 **2008**, *112*, 10823–10830.
30
31
32
- 33 (28) Prell, J. S.; Chang, T. M.; Biles, J. A.; Berden, G.; Oomens, J.; Williams, E. R.
34 Isomer Population Analysis of Gaseous Ions from Infrared Multiple Photon Dissociation
35 Kinetics. *The Journal of Physical Chemistry A* **2011**, *115*, 2745–2751.
36
37
38
- 39 (29) Polfer, N. C.; Oomens, J. Vibrational Spectroscopy of Bare and Solvated Ionic Com-
40 plexes of Biological Relevance. *Mass spectrometry reviews* **2009**, *28*, 468–494.
41
42
43
- 44 (30) Bleiholder, C.; Suhai, S.; Paizs, B. Revising the Proton Affinity Scale of the Natu-
45 rally Occurring α -amino Acids. *Journal of the American Society for Mass Spectrometry*
46 **2006**, *17*, 1275–1281.
47
48
49
50
51
52
53
54
55
56
57
58
59
60

- 1
2
3 (31) Francuski, B. M.; Novaković, S. B.; Bogdanović, G. A. Electronic Features and Hydro-
4 gen Bonding Capacity of the Sulfur Acceptor in Thioureido-based Compounds. Exper-
5 imental Charge Density Study of 4-methyl-3-thiosemicarbazide. *CrystEngComm* **2011**,
6 *13*, 3580–3591.
7
8
9
10
11
12 (32) Biswal, H. S.; Bhattacharyya, S.; Bhattacharjee, A.; Wategaonkar, S. Nature and
13 Strength of Sulfur-centred Hydrogen Bonds: Laser Spectroscopic Investigations in
14 the Gas Phase and Quantum-chemical Calculations. *International Reviews in Physi-*
15 *cal Chemistry* **2015**, *34*, 99–160.
16
17
18
19
20
21 (33) Aradi, B.; Hourahine, B.; Frauenheim, T. DFTB+, a Sparse Matrix-based Implementa-
22 tion of the DFTB Method. *The Journal of Physical Chemistry A* **2007**, *111*, 5678–5684.
23
24
25
26 (34) Density Functional Based Tight Binding (and more). <https://www.dftbplus.org/>,
27 Accessed 2019-02-12.
28
29
30
31 (35) Yatsyna, V.; Bakker, D. J.; Feifel, R.; Rijs, A. M.; Zhaunerchyk, V. Aminophenol Iso-
32 mers Unraveled by Conformer-specific far-IR Action Spectroscopy. *Physical Chemistry*
33 *Chemical Physics* **2016**, *18*, 6275–6283.
34
35
36
37 (36) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheese-
38 man, J. R.; Scalmani, G.; Barone, V.; Petersson, G. A.; Nakatsuji, H. et al. Gaussian~16
39 Revision C.01. 2016; Gaussian Inc. Wallingford CT.
40
41
42
43
44 (37) Halls, M. D.; Velkovski, J.; Schlegel, H. B. Harmonic Frequency Scaling Factors for
45 Hartree-Fock, S-VWN, B-LYP, B3-LYP, B3-PW91 and MP2 with the Sadlej pVTZ
46 Electric Property Basis Set. *Theoretical Chemistry Accounts* **2001**, *105*, 413–421.
47
48
49
50
51 (38) Johnson, E. R.; Keinan, S.; Mori-Sánchez, P.; Contreras-García, J.; Cohen, A. J.;
52 Yang, W. Revealing Noncovalent Interactions. *Journal of the American Chemical So-*
53 *ciety* **2010**, *132*, 6498–6506.
54
55
56
57
58
59
60

- 1
2
3 (39) Contreras-García, J.; Yang, W.; Johnson, E. R. Analysis of Hydrogen-bond Interaction
4 Potentials from the Electron Density: Integration of Noncovalent Interaction Regions.
5 *The Journal of Physical Chemistry A* **2011**, *115*, 12983–12990.
6
7
8
9
10 (40) Oh, H.-B.; Lin, C.; Hwang, H. Y.; Zhai, H.; Breuker, K.; Zabrouskov, V.; Carpen-
11 ter, B. K.; McLafferty, F. W. Infrared Photodissociation Spectroscopy of Electrosprayed
12 Ions in a Fourier Transform Mass Spectrometer. *Journal of the American Chemical So-*
13 *ciety* **2005**, *127*, 4076–4083.
14
15
16
17
18 (41) Adesokan, A. A.; Gerber, R. Anharmonic Vibrational Spectroscopy Calculations for
19 Proton-bound Amino Acid Dimers. *The Journal of Physical Chemistry A* **2008**, *113*,
20 1905–1912.
21
22
23
24
25 (42) Almasian, M.; Grzetic, J.; van Maurik, J.; Steill, J. D.; Berden, G.; Ingemann, S.;
26 Buma, W. J.; Oomens, J. Non-equilibrium Isomer Distribution of the Gas-phase Pho-
27 toactive Yellow Protein Chromophore. *The journal of physical chemistry letters* **2012**,
28 *3*, 2259–2263.
29
30
31
32
33 (43) Oomens, J.; Moore, D. T.; Meijer, G.; von Helden, G. Infrared Multiple Photon Dynam-
34 ics and Spectroscopy of Cationic PABA and its Dehydroxylated Fragment Ion. *Physical*
35 *Chemistry Chemical Physics* **2004**, *6*, 710–718.
36
37
38
39
40 (44) Maître, P.; Le Caër, S.; Simon, A.; Jones, W.; Lemaire, J.; Mestdagh, H.; Heninger, M.;
41 Mauclaire, G.; Boissel, P.; Prazeres, R. Ultrasensitive Spectroscopy of Ionic Reactive
42 Intermediates in the Gas Phase Performed with the First Coupling of an IR FEL
43 with an FTICR-MS. *Nuclear Instruments and Methods in Physics Research Section A:*
44 *Accelerators, Spectrometers, Detectors and Associated Equipment* **2003**, *507*, 541–546.
45
46
47
48
49 (45) Bouchoux, G. From the Mobile Proton to Wandering Hydride Ion: Mechanistic Aspects
50 of Gas-phase Ion Chemistry. *Journal of Mass Spectrometry* **2013**, *48*, 505–518.
51
52
53
54
55
56
57
58
59
60

- 1
2
3 (46) Poline, M.; Rebrov, O.; Larsson, M.; Zhaunerchyk, V. Theoretical Studies of Infrared
4 Signatures of Proton-bound Amino Acid Dimers with Homochiral and Heterochiral
5 Moieties. *Chirality* **2020**, *32*, 359–369.
6
7
8
9
10 (47) Headrick, J. M.; Diken, E. G.; Walters, R. S.; Hammer, N. I.; Christie, R. A.; Cui, J.;
11 Myshakin, E. M.; Duncan, M. A.; Johnson, M. A.; Jordan, K. D. Spectral Signatures
12 of Hydrated Proton Vibrations in Water Clusters. *Science* **2005**, *308*, 1765–1769.
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
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Graphical TOC Entry

