

Published in final edited form as:

Chemphyschem. 2009 October 19; 10(15): 2603–2606. doi:10.1002/cphc.200900564.

Numerous Isomers of Serine Octamer Ions Characterized by Infrared Photodissociation Spectroscopy

Xianglei Kong^[a], Cheng Lin^[a], Giuseppe Infusini^[a], Han-Bin Oh^[b], Honghai Jiang^[a], Kathrin Breuker^[c], Chih-Che Wu^[d], Oleg P. Charkin^[e], Huan-Cheng Chang^[f], and Fred W. McLafferty^[a]

^[a] Department of Chemistry and Chemical Biology Cornell University Ithaca, NY (USA) Fax: (+ 1) 607-25574137

^[b] Department of Chemistry, Sogang University Seoul, 200811036 (South Korea)

^[c] Institute of Organic Chemistry University of Innsbruck Innrain 52a, 6020 Innsbruck (Austria)

^[d] Department of Applied Chemistry National Chi Nan University Puli, Nantou (Taiwan)

^[e] Institute of Problems of Chemical Physics Russian Academy of Sciences Chernogolovka, Moscow 142432 (Russia)

^[f] Institute of Atomic and Molecular Sciences Academia Sinica, Taipei (Taiwan)

Keywords

amino acids; chirality; H/D exchange; infrared photodissociation spectroscopy; serine octamers

In contrast to several proposed mechanisms for prebiotic selection of L-amino acids in proteins, [1] formation of the gaseous magic-number cluster $(\text{Ser}_8+\text{H})^+$ achieves extensive homochirality; D,L-Ser selectively yields $(\text{D-Ser}_8+\text{H})^+$ and $(\text{L-Ser}_8+\text{H})^+$. [2] Using processes that simulate prebiotic conditions, serine octamers are also selectively formed as neutrals and anions and from solid and solution phases. $(\text{Ser}_8+\text{H})^+$ is far more stable than homochiral octamers of any other of the 20 naturally-occurring amino acids (X), and single substitutions of the latter into $(\text{L-Ser}_8+\text{H})^+$ are enantioselective to form $(\text{L-Ser}_{7-\text{L-X}}+\text{H})^+$. However, high chiral selectivity of an enzyme is usually found to result from a unique binding-site conformation, while recent infrared photodissociation (IRPD) spectra [3] from our Cornell and Taiwan laboratories found contrasting structures containing no [4] and multiple [5] free aliphatic –OH groups, respectively. Herein we reexamine $(\text{Ser}_8+\text{H})^+$ IRPD spectra from ions formed under a variety of conditions.

$(\text{Ser}_8+\text{H})^+$ ions have been extensively studied by experimental [1–9] and theoretical [6b,7,8] methods. Although all $(\text{Ser}_8+\text{H})^+$ ions studied have nearly identical cross sections ($\sim 190 \text{ \AA}^2$), [6b,7] two isomeric forms have been distinguished by their H/D exchange rates. [2c,9] The faster-exchanging isomer B shows no chiral selection and is selectively dissociated [9] in collisional SWIFT isolation [10] of the A/B mixture. [2,9] Theory predicts a zwitterionic structure for isomer A, with the free (i.e. without H-bonding) aliphatic O–H group absent from that of Beauchamp et al. [6b] and present in the Schalley et al. structure. [8]

IRPD spectra[3] in the O–H/N–H stretching region were measured on electrosprayed (Ser_8+H)⁺ ions (Figure 1) stored in a Fourier-transform mass spectrometer.[4,5] The Cornell 2003 spectrum[4,11] (as well as pre-2006 spectra, not shown, measured by Oh in Korea) did not absorb at $\sim 3675\text{ cm}^{-1}$ (isomer *i*, Table 1), indicating the absence of free aliphatic O–H groups. However, this peak was strong in the 2006 Taiwan spectrum (isomer *ii*).[5] Negligible absorption at $\sim 3565\text{ cm}^{-1}$ (Figure 1) shows the absence of free carboxyl O–H,[4,5] supporting a zwitterionic structure. Some Cornell 2004 spectra (Figure 1) do have a new absorption at 3565 cm^{-1} that indicates a new isomer *iii* (Table 1).

Additional Cornell IRPD spectra (termed “2007”) were measured under what was thought to be the same experimental conditions, but without SWIFT isolation that removes isomer B.[9] Although our 2007 spectra are quite similar, now the 3675 cm^{-1} peak has one-third the relative intensity of the corresponding Taiwan 2006 peak. This decrease is not due to a $\sim 1:2$ mixture of isomer *ii* with *i* (which does not absorb at 3675 cm^{-1}), because IR irradiation at this frequency causes (Figure 2) essentially complete dissociation of these 2007 ions without forming *i*. The data in Figure 2 show a first-order correlation, indicating that both 2007 forms A and B (Figure 3 a, without SWIFT isolation) exhibit similar dissociation kinetics over this 200 s time period. The 2007 ions from SWIFT isolation, presumably isomer *iv*, gave an IRPD spectrum whose main features were closely similar to those of the *iv/v* mixture.

In contrast to these equivalent A/B photodissociation rates, initial SWIFT isolation of the 2007 ions (Figure 3 b), in which both the A and B forms at 10^{-9} torr are translationally excited by off-resonance frequencies followed after 25 s by a collisionally-activating gas pulse, reduces the intensity of B to a far lower value ($\sim 10\%$) than that of A ($\sim 60\%$).[2c,9] In further apparent contrast, conventional collisional activation (sustained off-resonance irradiation)[12] that uses far faster (~ 0.1 s), higher-energy collisions (Figure 3 c) gave more comparable values: 46% for A and 33% for B (as shown by Figure 3 d, this is not due to the isomerization of A to B). Further, for the IRPD spectrum (not shown) of these product ions, the 3675 cm^{-1} free O–H absorption has been dramatically reduced to $<10\%$ of its original value (Figure 4), with that at 3325 cm^{-1} halved. Thus, the spectrum is closely similar to that of isomer *i*, suggesting *i* as a collisional activation product of the 2007 ions. Its B form counterpart is then the new isomer *vi* (Table 1; 2003 ions did not contain the B form).

Possible reactions of these A and B isomers are given by Equations (1)–(3), respectively:

Dissociation :



;



. Isomerization :



;



. Dissociation :



;



At the low energy of Figure 2, the dissociation rates of Equations (1a) and (1b) are equivalent, but at the higher-energy SWIFT excitation (Figure 3 b) reaction (1b) is substantially faster, indicating a looser transition state. However, both (1a) and (1b) could involve an initial tight-complex isomerization, as even looser transition states are indicated for the those of Equations (2a) and (2b) that become competitive when activated at the highest Figure 3 c energy. Here their rates are greater than those of dissociations (3a) and (3b) [as well as (1a) and (1b)], indicating that isomers *i* and *vi* (no free O–H) are substantially more stable than *iv* and *v*. Similarly, *ii* with the most free O–H groups could be even less stable, avoiding isomerization to *i* or *iv* only under gentle ion introduction conditions.[5] Quite different H/D exchange rates have also been noted for other gaseous conformers of similar structure.[3a,13] The A forms *ii* and *iii* could have B counterparts *vii* and *viii*, while other stable A forms could give similar IRPD spectra.

Of the $(\text{Ser}_8+\text{H})^+$ structures proposed previously from theory, the zwitterionic form I (Figure 5) of Beauchamp et al.[6b] has been found by Schalley to be the most favorable.[8] Further calculations[14] here found at least seven stable isomers. The most stable (I, II, III) contain 0, 1, and 6 free aliphatic O–H groups, respectively, as reflected in the relative intensities of the 3675 cm^{-1} peaks in their computed IR spectra (Figure 5), with relative energies of 0, -13.5 , and -11.9 kJ mol^{-1} . However, I–III only reflect general properties of the Table 1 isomers *i*–*vi*, not specific characteristics. The absence of a free O–H peak from structure I is consistent with isomers *i*, *iii*, and *vi*, the weak peak of II with *iv* and *v*, and the strong peak of III with *ii*. However, *i* and *vi* (supposedly like I) are produced by activation of *iv* and *v* (II) by Equations (2a) and (2b), inconsistent with the lower relative energy of II. The 3565 cm^{-1} peaks in I and II are due to $\text{CH}_2\text{O}-\text{H}\cdots(\text{OCO})^-$ (not free $\text{COO}-\text{H}$), suggesting this assignment for isomer *iii* as well. No free $\text{COO}-\text{H}$ in any of I–III suggests that all have zwitterionic structures.[15]

In summary, experiment and theory indicate at least six stable $(\text{Ser}_8+\text{H})^+$ isomers.[16] these have eight aliphatic O–H groups variously H-bonded around a zwitterionic core. However, only kinetic dissociation data [Eq. (1a)–(3b)], not IRPD spectral features, distinguish between the enantiomerically active and inactive forms A and B. A singular conformational structure is often cited as basic for an enzyme's unusually high specificity, but here the specificity of serine octamer is also of unusually broad applicability, as it acts for all the essential l-amino acids (X) to select $l\text{-X}$, not $d\text{-X}$, in $(l\text{-Ser}_7\text{-l-X}+\text{H})^+$. The structural variability of the –OH groups on the outside of the cluster could optimize the conformation for substituting any amino acid for a single serine while retaining the unique enantioselectivity of its singular zwitterionic center.

Experimental Section

IRPD spectra of $(\text{Ser}_8+\text{H})^+$ ions were obtained at Cornell University as described previously [4,11] using a 6T Fourier-transform MS; its resolving power of $\sim 10^5$ shows no more highly charged ions such as $(\text{Ser}_{16}+2\text{H})^{2+}$. Ions from nano-electrospray ionization of 5 mM L-serine (Sigma, St. Louis) in 49:49:2 MeOH:H₂O:AcOH were trapped in the ion cell (N₂ pulse,

$\sim 10^{-6}$ Torr). Where noted, all but the $(\text{Ser}_8+\text{H})^+$ ions were ejected with a SWIFT waveform [10] at 10^{-9} Torr, followed 25 s later by a 10^{-6} torr N_2 pulse. Sustained off-resonance irradiation [12] used 38 db, -1 kHz off resonance, for 0.1 s at $\sim 10^{-6}$ Torr N_2 . H/D exchange used CH_3OD in the FTMS cell at 6×10^7 Torr for 50 s, then a 300 s pump down.

IRPD spectra were measured at $\sim 10^{-9}$ Torr with a pulsed infrared laser ($3025\text{--}3775\text{ cm}^{-1}$, IR OPO 2732, OPOTek, Carlsbad, CA) pumped by a ND:YAG laser, output 4–9 mJ at 10 Hz. Intensities of the $(\text{Ser}_8+\text{H})^+$ ions, relative to the laser power, were determined before and after photodissociation. Irradiation time at each frequency, typically 15 s, was controlled by a mechanical shutter (UNI-BLITZ, Vincent Assoc., Rochester, NY).

Theoretical calculations at the Computer Center, Academia Sinica, used the Gaussian 03 program package.[14] All structures and frequency calculations were optimized by DFT at the B3LYP/6-31G(d) level. Final energies were refined by B3LYP/6-311 ++ G(d,p) single-point calculations and B3LYP/6-31G(d) zero-point energy corrections. IR spectra were calculated at B3LYP/6-31G(d) and scaled with a single factor of 0.97.

Acknowledgments

We thank to Drs. J. L. Beauchamp, S. Castro, R. G. Cooks, J. C. Jiang, and C. A. Schalley for helpful discussions and the Academia Sinica and the National Institute of General Medical Science, NIH (GM16609) for financial support.

References

1. Breslow R, Cheng Z-L. Proc. Natl. Acad. Sci. USA 2009;106:9144–9146. [PubMed: 19478058]and references therein
2. a Nanita SC, Cooks RG. Angew. Chem 2006;118:568–583. Angew. Chem. Int. Ed 2006;45:554–569. b Cooks RG, Zhang D, Koch KJ, Gozzo FC, Eberlin MN. Anal. Chem 2001;73:3646–3655. [PubMed: 11510829] c Takats Z, Nanita SC, Schlosser G, Vekey K, Cooks RG. Anal. Chem 2003;75:6147–6154. [PubMed: 14615994]
3. a Oh H-B, Breuker K, Sze SK, Ge Y, Carpenter BK, McLafferty FW. Proc. Natl. Acad. Sci. USA 2002;99:15863–15868. [PubMed: 12444260] b Polfer NC, Oomens J. Mass Spectrom. Rev 2009;28:468–494. [PubMed: 19241457]
4. Hwang, HY.; Lin, C.; Oh, H.; Breuker, K.; Carpenter, BK.; McLafferty, FW. Proceedings, 52nd ASMS Conference on Mass Spectrometry; Nashville, TN, USA. 2004; b Oh HB, Lin C, Hwang HY, Zhai H, Breuker K, Zabrouskov V, Carpenter BY, McLafferty FW. J. Am. Chem. Soc 2005;127:4076–4083. [PubMed: 15771545]
5. Kong X, Tsai I-A, Sabu S, Han CC, Lee YT, Chang HC, Tu S-Y, Kung AH, Wu C-C. Angew. Chem 2006;118:4236–4240. Angew. Chem. Int. Ed 2006;45:4130–4134.
6. a Hodyss R, Julian RR, Beauchamp JL. Chirality 2001;13:703–706. [PubMed: 11746805] b Julian RR, Hodyss R, Kinnear B, Jarrold MF, Beauchamp JL. J. Phys. Chem. B 2002;106:1219–1228.
7. Counterman AE, Clemmer DE. J. Phys. Chem. B 2001;105:8092–8096.
8. Schalley CA, Weis P. Int. J. Mass Spectrom 2002;221:9–19.
9. Mazurek U, Geller O, Lifshitz C, McFarland MA, Marshall AG, Reuben BG. J. Phys. Chem. A 2005;109:2107–2112. [PubMed: 16838981]
10. Marshall AG, Wang TCL, Ricca TL. J. Am. Chem. Soc 1985;107:7893–7897.
11. The apparent frequency discrepancy between spectra from the three laboratories[4,5] was resolved by recalibration of the Cornell and Korea OPO lasers. All IRPD frequencies of reference [4] should be corrected by -25 cm^{-1} , and have been so corrected herein
12. Gauthier JW, Trautman TR, Jacobson DB. Anal. Chim. Acta 1991;246:211–225. Senko MW, Speir JP, McLafferty FW. Anal. Chem 1994;66:2801–2808. [PubMed: 7978294]
13. a McLafferty FW, Guan Z, Happts U, Wood TD, Kelleher NL. J. Am. Chem. Soc 1998;120:4732–4740. b Freitas MA, Hendrickson CL, Emmett MR, Marshall AG. Int. J. Mass Spectrom 1999;185/186/187:565–575.

14. Frisch, MJ.; Trucks, GW.; Schlegel, HB.; Scuseria, GE.; Robb, MA.; Cheeseman, JR.; Montgomery, JA., Jr.; Vreven, T.; Kudin, KN.; Burant, JC.; Millam, JM.; Iyengar, SS.; Tomasi, J.; Barone, V.; Mennucci, B.; Cossi, M.; Scalmani, G.; Rega, N.; Petersson, GA.; Nakatsuji, H.; Hada, M.; Ehara, M.; Toyota, K.; Fukuda, R.; Hasegawa, J.; Ishida, M.; Nakajima, T.; Honda, Y.; Kitao, O.; Nakai, H.; Klene, M.; Li, X.; Knox, JE.; Hratchian, HP.; Cross, JB.; Bakken, V.; Adamo, C.; Jaramillo, J.; Gomperts, R.; Stratmann, RE.; Yazyev, O.; Austin, AJ.; Cammi, R.; Pomelli, C.; Ochterski, JW.; Ayala, PY.; Morokuma, K.; Voth, GA.; Salvador, P.; Dannenberg, JJ.; Zakrzewski, VG.; Dapprich, S.; Daniels, AD.; Strain, MC.; Farkas, O.; Malick, DK.; Rabuck, AD.; Raghavachari, K.; Foresman, JB.; Ortiz, JV.; Cui, Q.; Baboul, AG.; Clifford, S.; Cioslowski, J.; Stefanov, BB.; Liu, G.; Liashenko, A.; Piskorz, P.; Komaromi, I.; Martin, RL.; Fox, DJ.; Keith, T.; Al-Laham, MA.; Peng, CY.; Nanayakkara, A.; Challacombe, M.; Gill, PMW.; Johnson, B.; Chen, W.; Wong, MW.; Gonzalez, C.; Pople, JA. Gaussian 03, Revision B.01. Gaussian, Inc.; Wallingford CT: 2004.
15. For the characteristic $\sim 3325\text{ cm}^{-1}$ absorption of Table 1, $\text{N}^+\text{H}_2\text{—H}$ or symmetric $\text{H—N}^+\text{—H}$ stretches are predicted in the IR spectra of I ($3320, 3360, 3400\text{ cm}^{-1}$) and III ($3295, 3325, 3355\text{ cm}^{-1}$), but the 3340 cm^{-1} peak of II arises from H-bonded $\text{CH}_2\text{O—H}$
16. Suckau D, Shi Y, Beu SC, Senko MW, Wampler FM, McLafferty FW. Proc. Natl. Acad. Sci. USA 1993;90:790–793. [PubMed: 8381533] Numerous isomers are common for other gaseous ions: e.g. Koeniger SL, Merenbloom SI, Clemmer DE. J. Phys. Chem. B 2006;110:7017–7021. [PubMed: 16571016] Breuker K, McLafferty FW. Proc. Natl. Acad. Sci. USA 2008;105:18145–18152. [PubMed: 19033474]

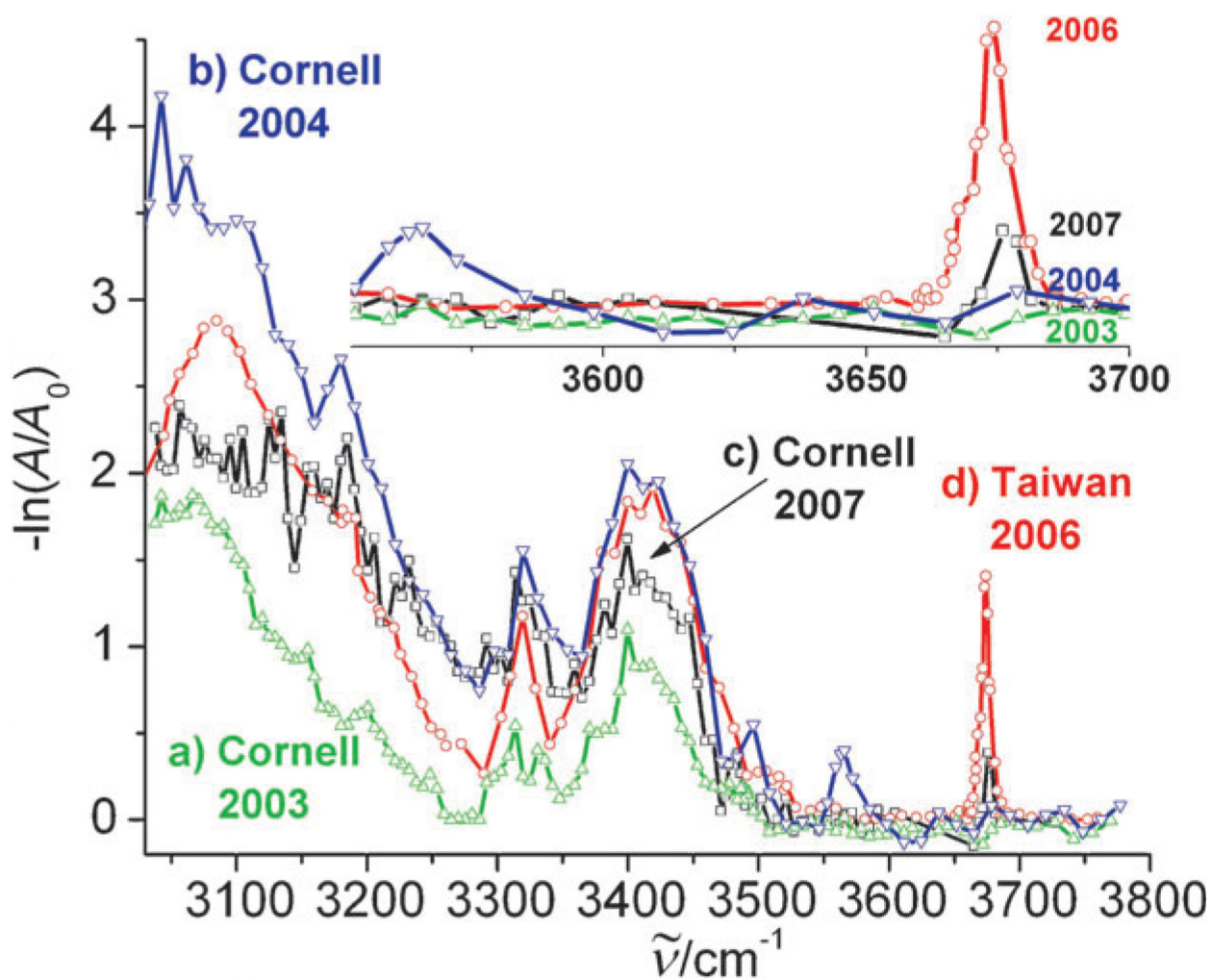


Figure 1. IRPD spectra of $(L\text{-Ser}_8\text{+H})^+$ measured at Cornell in about a) 2003,[4,11] b) 2004 [racemic $(\text{Ser}_8\text{+H})^+$ ions measured in 2004 gave a very similar spectrum], c) 2007, and in Taiwan reported[5] in d) 2006. Only the 2007 ions were not SWIFT-isolated and so should contain the isomeric form B.

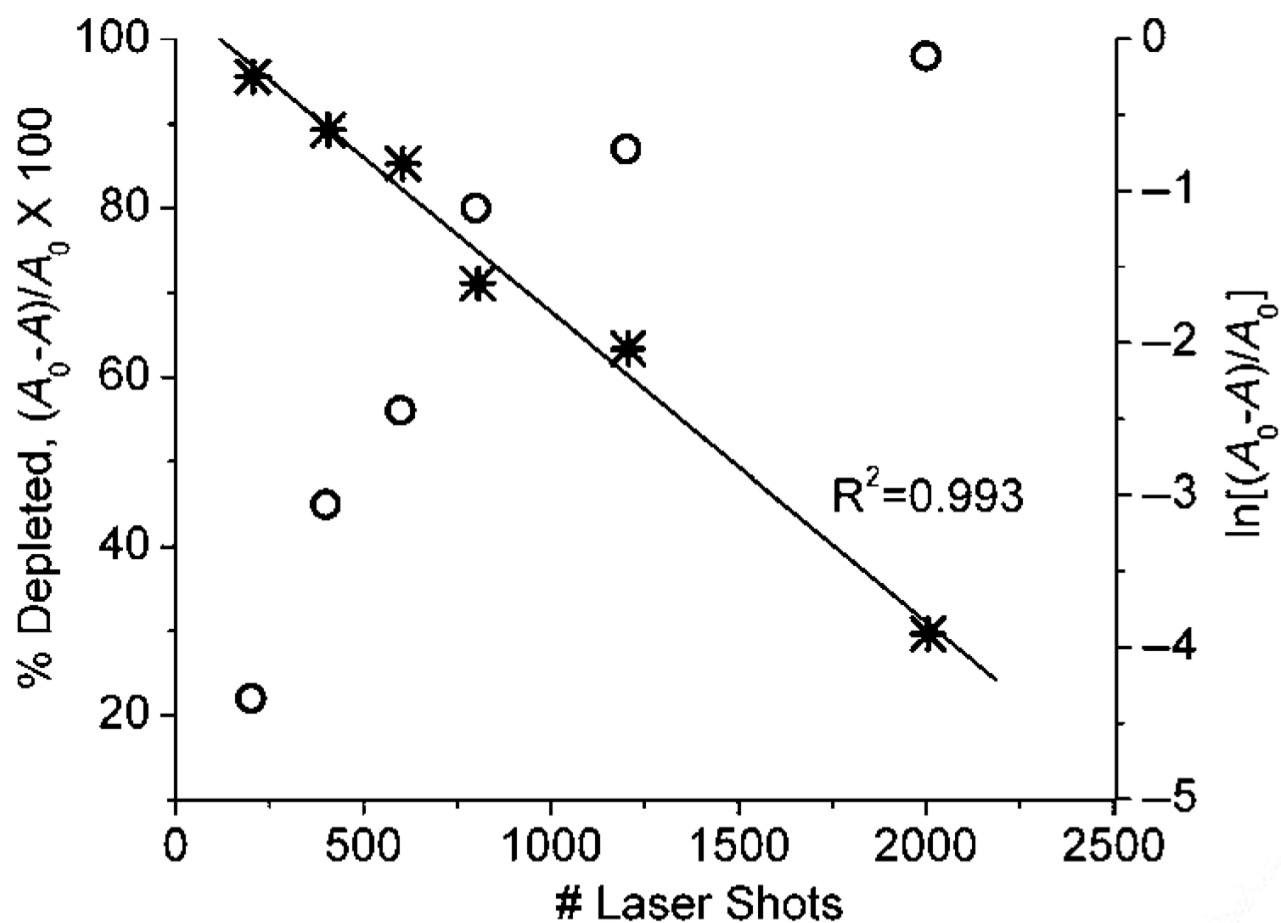


Figure 2. Photodissociation of 2007 (Ser₈+H)⁺ ions (not SWIFT-isolated): (○), left Y axis, abundance depletion versus the number of 3676 cm⁻¹ laser pulses, ~4.5 mJ/pulse, 10 Hz; *, right Y-axis, log relative abundance. The correlation line as drawn indicates that dissociation onset requires ~100 laser shots.

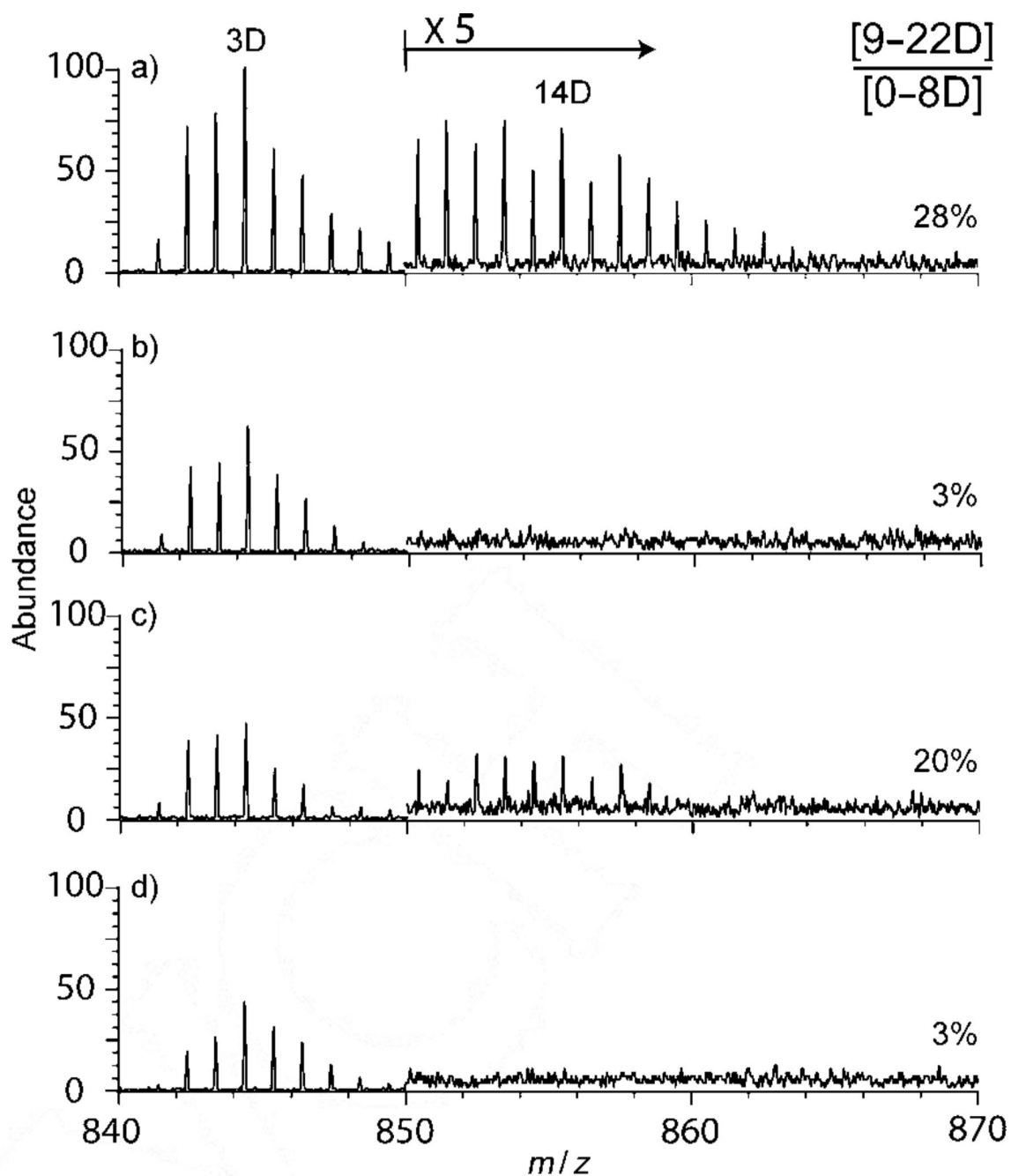


Figure 3. Gas-phase H/D exchange of 2008 (Ser8+H)⁺ ions a) as trapped in the FTMS cell, b) after their SWIFT isolation, c) after their collisional activation, and d) after the same SWIFT isolation and collisional activation (lower intensity, 39 db) of these ions.

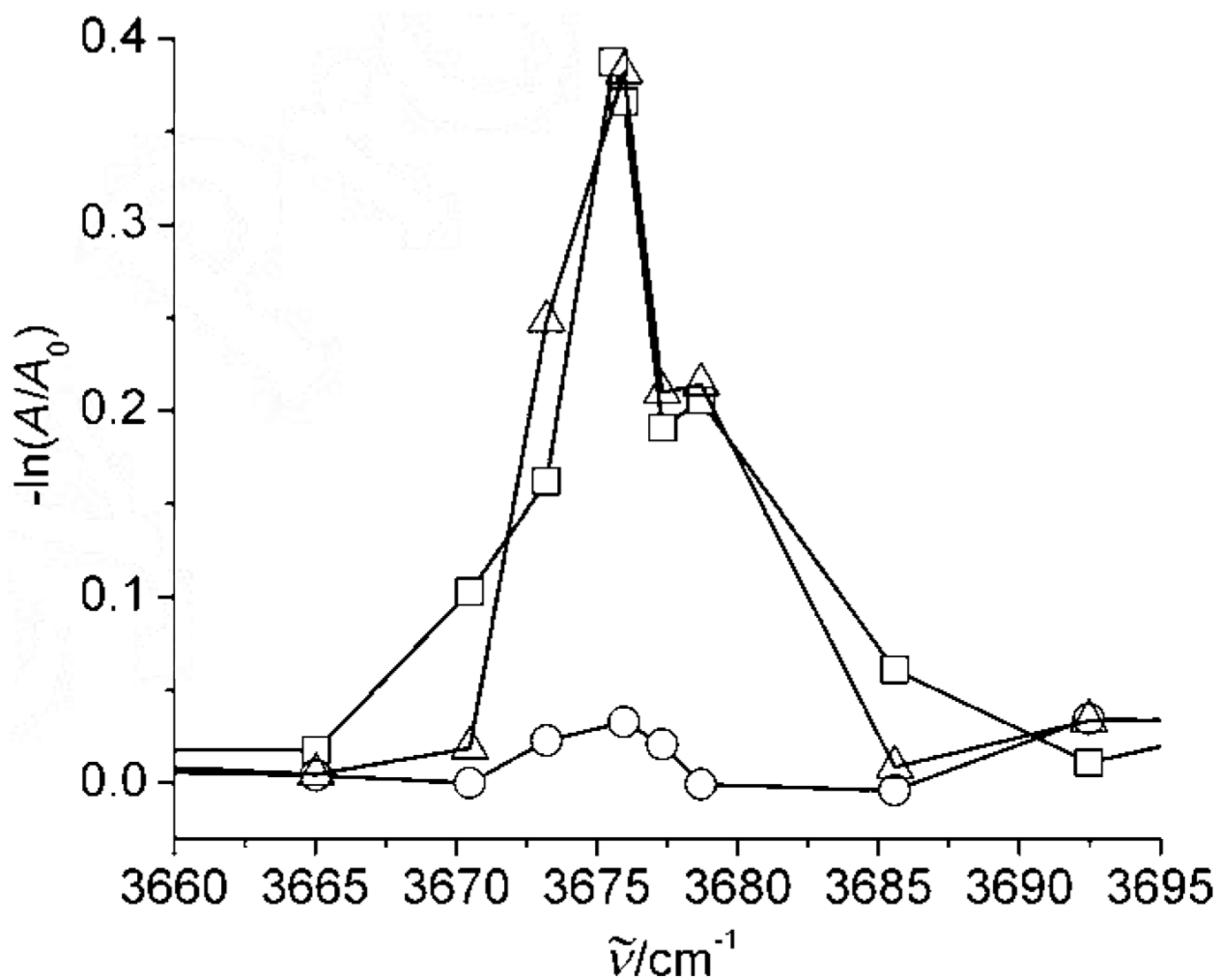


Figure 4. Partial IRPD spectra of 2007 $(\text{Ser}_8+\text{H})^+$ ions: (\square) as electrosprayed, (Δ) after SWIFT isolation, (\circ) after collisional activation. These treatments resemble those of Figures 3 a–c, but without H/D exchange.

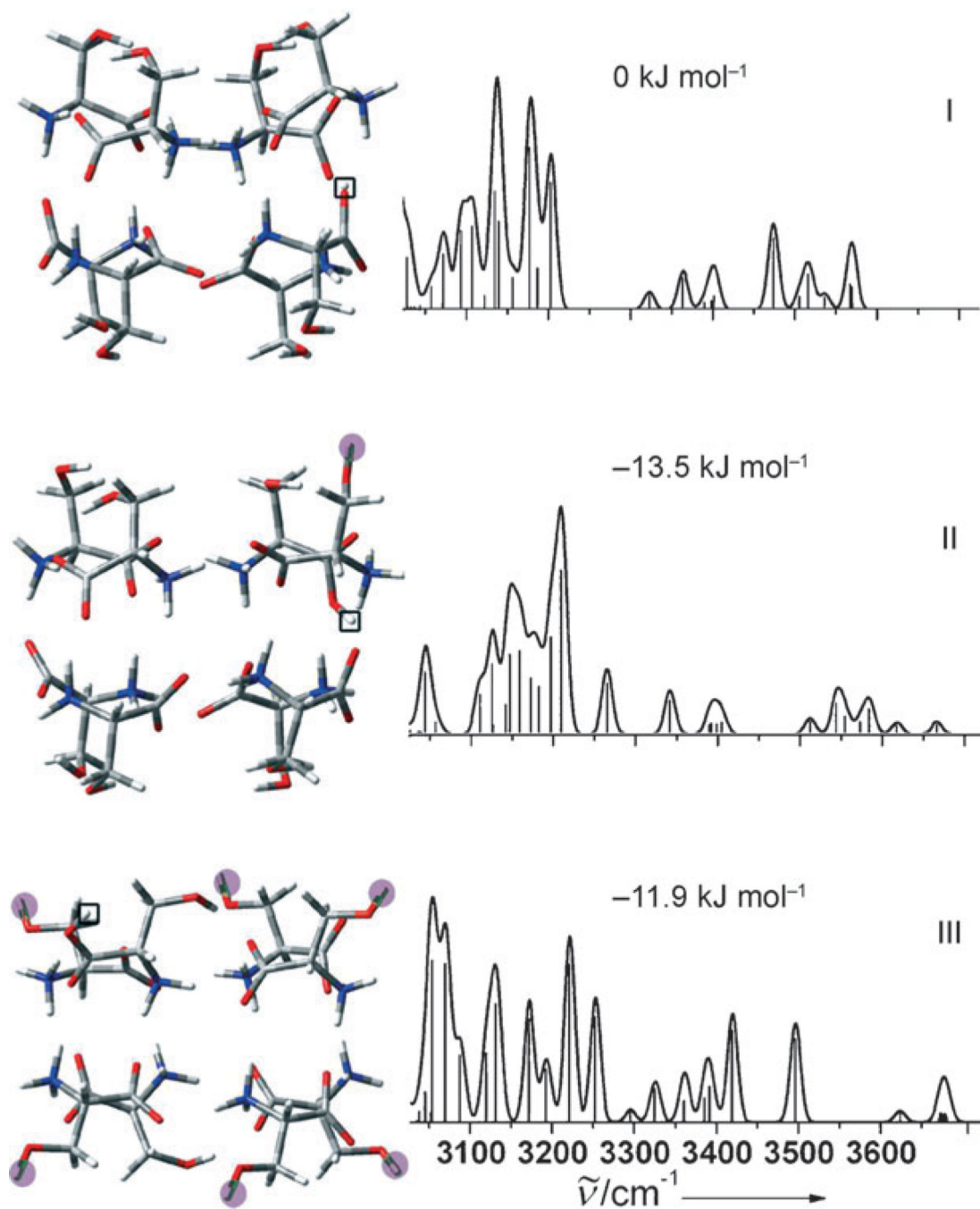


Figure 5. Predicted structures and IR spectra of $(\text{Ser}_8+\text{H})^+$; I is based on that of Beauchamp et al.[6b] Shaded circle: free aliphatic O-H group (0, 1, and 6 groups in I, II, and III, respectively); open square, the proton.

Table 1

(Ser₈+H)⁺ ion properties versus possible isomers.

Major isomer	Spectrum	H/D exch. isomer	IRPD absorption [cm ⁻¹]	3675	3565	3325
<i>i</i>	2003[4]	A ^[a]	-	-	-	+
<i>ii</i>	2006[5]	A	+++	-	-	++
<i>iii</i>	2004	A ^[a]	-	+	+	++
<i>iv</i>	2007	A	+	-	-	++
<i>v</i>	2007	B	+	-	-	++
<i>i</i>	col. activ. 2007 ^[b]	A	-	-	-	+
<i>vi</i>	col. activ. 2007 ^[b]	B	-	-	-	+

^[a] Ions isolated by SWIFT, so isomeric form B presumably was lost.[9]

^[b] For these isomerization products, the IRPD spectrum resembles that of the *i* isomer, so that the B form should be new isomer *vi*.