

On the turn-inducing properties of asparagine: the structuring role of the amide side chain, from isolated model peptides to crystallized proteins

S. Habka,^a, W.Y. Sohn,^{a,†} V. Vaquero-Vara,^a M. Géléoc,^a B. Tardivel,^a V. Brenner,^a E. Gloaguen,^a M. Mons^a

LIDYL; CEA, CNRS, Université Paris Saclay; CEA Saclay, Bât 522, 9119 Gif-sur-Yvette, France

[†] Present address: Department of Applied Chemistry, Faculty of Science and Technology, Chuo University, 1-13-27 Kasuga, Bunkyo, Tokyo 112-8551, Japan

Supplementary information

Figure S1 : Near UV spectrum of jet-cooled Z-Asn-NH₂

Table S1 : Conformer-selective IR spectroscopy of the Z-Asn-NH₂ molecule

Table S2 : Conformer-selective IR spectroscopy of the Ac-Phe-Asn-NH₂ molecule

Table S3 : Conformer-selective IR spectroscopy of the Ac-Asn-Phe-NH₂ molecule

Figure S2 : Distribution of side chain orientations for gauche+ Asp residues in position i+2 of a β-turn, for the subpopulation of turns exhibiting an elongated 6^δ interaction

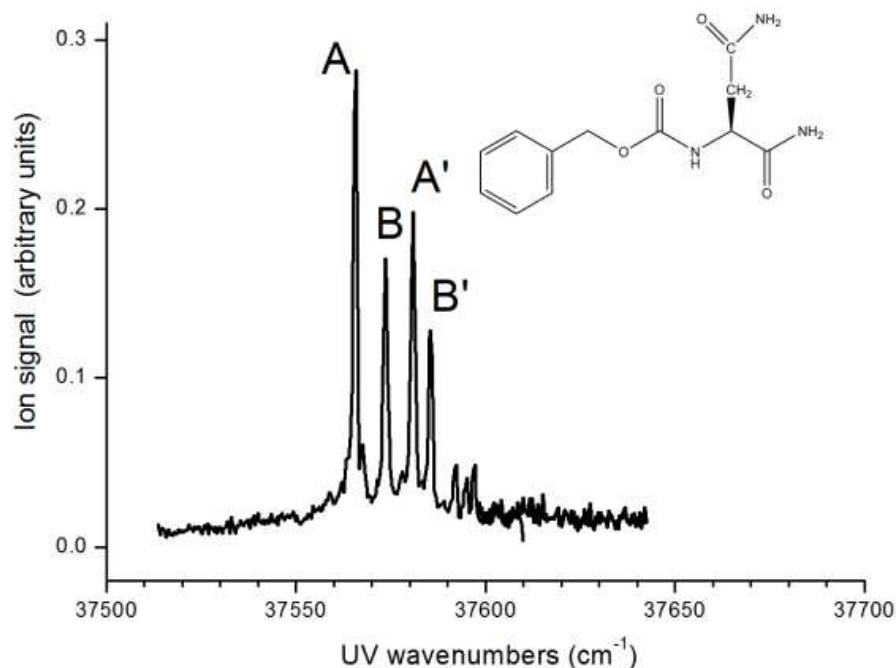


Figure S1 : Near UV spectrum of jet-cooled Z-Asn-NH₂, in the origin region of the first electronic transition of the phenyl ring, as obtained by mass-selected resonant two-photon ionization spectroscopy. The IR/UV double resonance spectra carried out by probing the most intense bands observed has allowed us to determine the existence of two conformers and the corresponding two sets of UV transitions (A-A' and B-B').

Table S1 : Conformer-selective IR spectroscopy of the Z-Asn-NH₂ molecule : Experimental NH stretch frequencies (cm⁻¹) of the two conformers (A & B) detected in the supersonic expansion, compared to the predicted frequencies of extended and folded conformations for this molecule and the simple Ac-Asn-NH₂ model molecule. Also given are, the 0K and 300 K relevant energetics of these conformations (kJ/mol), two indicators of the error between predicted spectra and observed conformers' frequencies (δ^{ave} , the average RMS error, and δ^{max} , the unsigned maximum error). The best fits according to frequency (small δ^{ave} and δ^{max} values) and energetic criteria (small ΔG values) are indicated in bold. Predicted frequencies are obtained at the B97-D3/TZVPP level of theory, by applying a mode-dependent two-parameter scaling factor to the harmonic NH stretch frequencies. For all these molecules, only the most stable form of the benzyloxycarbonyl (Z-) cap, i.e., its extended conformation (N(i)C(i+1)OC_{carb.} ~180°), has been considered.

Z-Asn-NH₂ Experimental frequencies											
	A										
	B										
Z-Asn-NH₂ Theoretical frequencies											
Conformation	short and full names	assignment	ΔH	ΔG 300 K	NH ₂ ^{anti}	NH ₂ ^{sym}	NH _{Asn}	NH ₂ ^{anti} Asn sidechain	NH ₂ ^{sym} Asn sidechain	A $\delta^{ave}/\delta^{max}$	B $\delta^{ave}/\delta^{max}$
a	6^δ-7 (δ⁷)	A	1	1	3514	3331	3390	3510	3387	10/27	18/33
b	5-7^δ (δ⁸)	B	0	0	3516	3320	3416	3518	3390	16/27	7/14
Ac-Asn-NH₂ Theoretical frequencies											
a	6 ^δ -7 (δ ⁷)		0	0	3511	3301	3380	3511	3388		
b	5-7 ^δ (δ ⁸)		1	0	3515	3318	3410	3515	3379		

Table S2 : Conformer-selective IR spectroscopy of the Ac-Phe-Asn-NH₂ molecule; same details as Table S1. The Phe side chain orientation is gauche+ in the lowest conformation for each backbone considered.

Ac-Phe-Asn-NH₂													
Experimental frequencies													
A		3369; 3389; 3418; 3442; 3517; 3533											
B		3312; 3328; 3389; 3451; 3503; 3511											
C		3305; 3362; 3391; 3456; 3513; 3521											
Ac-Phe-Asn-NH₂													
Theoretical frequencies													
Conformation short and full names		assignment	ΔH	ΔG 300 K	NH ₂ _{anti}	NH ₂ _{sym}	NH Phe	NH Asn	NH ₂ _{anti} Asn sidechain	NH ₂ _{sym} Asn sidechain	A δ ^{ave} / δ ^{max}	B δ ^{ave} / δ ^{max}	C δ ^{ave} / δ ^{max}
a	π ^F -6 ^δ -10 (δ ⁷)	A	0	3.2	3520	3377	3439	3411	3523	3408	8/19	23/45	22/47
b	5-6 ^δ /π ^F -7 (δ ⁷)	B	2.5	2.8	3506	3324	3447	3369	3513	3379	34/80	11/36	12/42
c	5-5/π ^F -7 ^δ (δ ⁸)	C	2.3	0	3516	3387	3452	3364	3515	3322	29/76	6/22	11/24
Ac-Ala-Asn-NH₂													
Theoretical frequencies													
a	π ^F -6 ^δ -10 (δ ⁷)				3520	3376	3472	3409	3523	3408			

Table S3 : Conformer-selective IR spectroscopy of the Ac-Asn-Phe-NH₂ molecule; same details as Table S1. The Phe side chain orientation is gauche+ in the lowest conformation for each backbone considered.

Ac-Asn-Phe-NH₂												
Experimental frequencies												
A		3381(broad); 3414; 3430; 3520; 3545										
B		3367; 3391 (broad); 3455; 3511; 3519										
Ac-Asn-Phe-NH₂												
Theoretical frequencies												
Conformation short and full names		assignment	ΔH	ΔG 300 K	NH ₂ _{anti}	NH ₂ _{sym}	NH Asn	NH Phe	NH ₂ _{anti} Asn sidechain	NH ₂ _{sym} Asn sidechain	A ^a δ ^{ave} / δ ^{max}	B ^a δ ^{ave} / δ ^{max}
a	6 ^δ -π ^F -10 (δ ⁷)	A	0.7	0	3531	3386	3400	3450	3531	3417	11/21	18/42
b	6 ^δ -f-10 (δ ⁷ /δ ^π)	B	0	3.1	3523	3389	3389	3471	3523	3396	14/26	8/22
Ac-Asn-Ala-NH₂												
Theoretical frequencies												
a	6 ^δ -f-10 (δ ⁷)				3520	3388	3391	3479	3530	3413		

a) assuming that the broad bands result from overlaps of two adjacent bands.

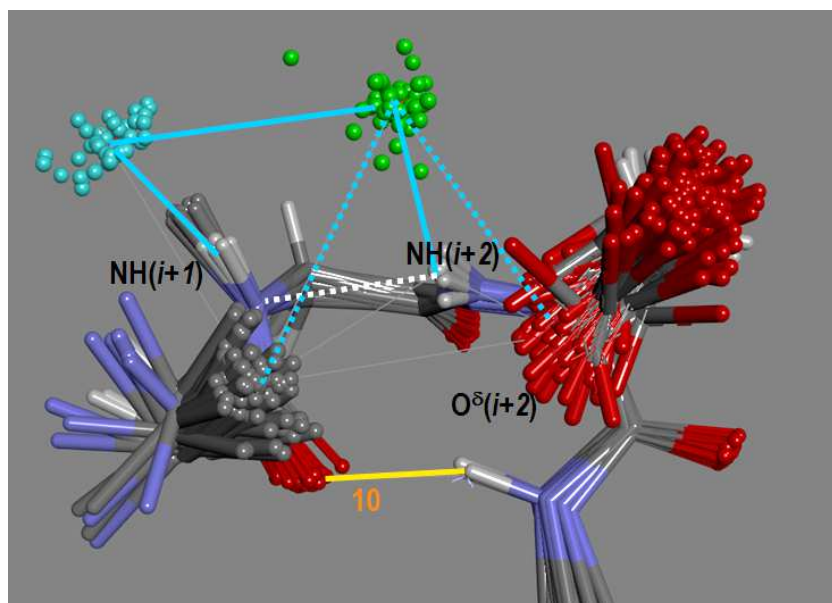


Figure S2 : Distribution of side chain orientations for gauche+ Asp residues in position $i+2$ of a β -turn, for the subpopulation of turns exhibiting with an elongated 6^δ interaction, obtained from a sampling of high resolution X-ray structures (resolution lower than 1.6\AA) in the PDB . Same caption details as Figure 9. The oxygen atoms of hydration molecules detected in the crystals are visualized as small balls, coloured as a function of their spatial location; they form cluster-like groups, providing evidence for the existence of a robust hydration site.