SUPPORTING INFORMATION

Effect of 3-Hydroxyproline Residues on Collagen Stability

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Table S1. Data for pH Titration of 3-Hydroxyproline

Sample	pH	δ (α-proton, Hz)	Volume (μL) of 0.10 M D_3PO_4	Volume (μL) of 0.10 M NaOD	Volume (μL) of 0.10 M DCl
1	1.30	2129.1	950	0	0
2	1.36	2124.2	902	48	0
3	1.42	2118.3	855	95	0
4	1.50	2111.2	807	143	0
5	1.57	2104.6	760	190	0
6	1.66	2096.2	712	238	0
7	1.78	2087.5	665	285	0
8	1.93	2077.7	617	333	0
9	2.10	2067.9	570	380	0
10	2.39	2053.5	522	428	0
11	3.05	2039.4	475	475	0
12	0.61	2166.8	0	0	950
13	0.73	2162.1	237	0	713
14	0.86	2155.5	475	0	475
15	1.05	2144.5	713	0	237

Table S2. Data for pH Titration of 4-Hydroxyproline

Sample	pH	δ (α-proton, Hz)	Volume (µL) of 0.10 M D_3PO_4	Volume (µL) of 0.10 M NaOD	Volume (µL) of 0.10 M DCl
1	1.33	2290.8	950	0	0
2	1.39	2286.5	902	48	0
3	1.47	2280.5	855	95	0
4	1.52	2272.8	807	143	0
5	1.62	2264.0	760	190	0
6	1.71	2254.0	712	238	0
7	1.84	2244.5	665	285	0
8	1.99	2228.0	617	333	0
9	2.17	2214.9	570	380	0
10	2.48	2195.0	522	428	0
11	3.31	2173.1	475	475	0
12	0.60	2325.1	0	0	950
13	0.70	2321.6	237	0	713
14	0.86	2315.0	475	0	475
15	1.02	2305.2	713	0	237

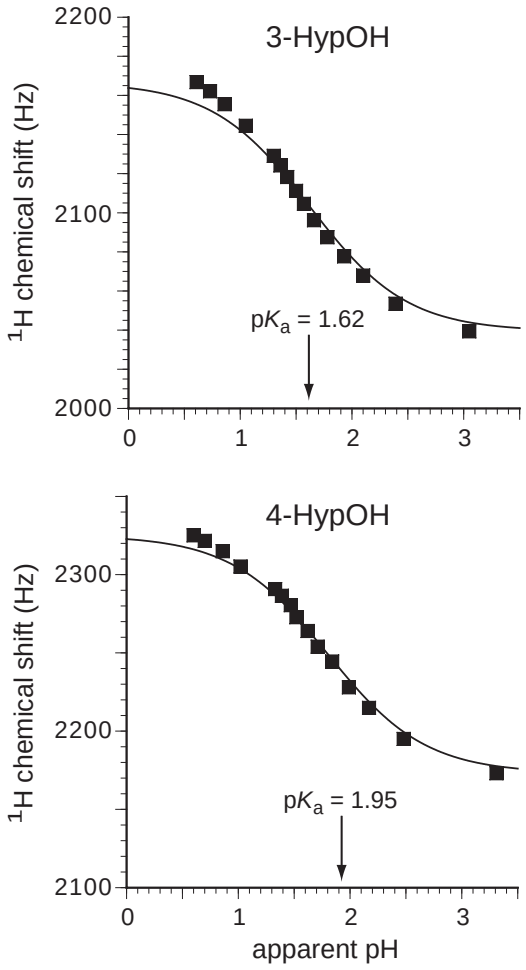

Figure S1. Effect of pH on the ^{1}H NMR chemical shift of 3-HypOH and 4-HypOH. Data were fitted with eq 1 to give the indicated values of pK_a.

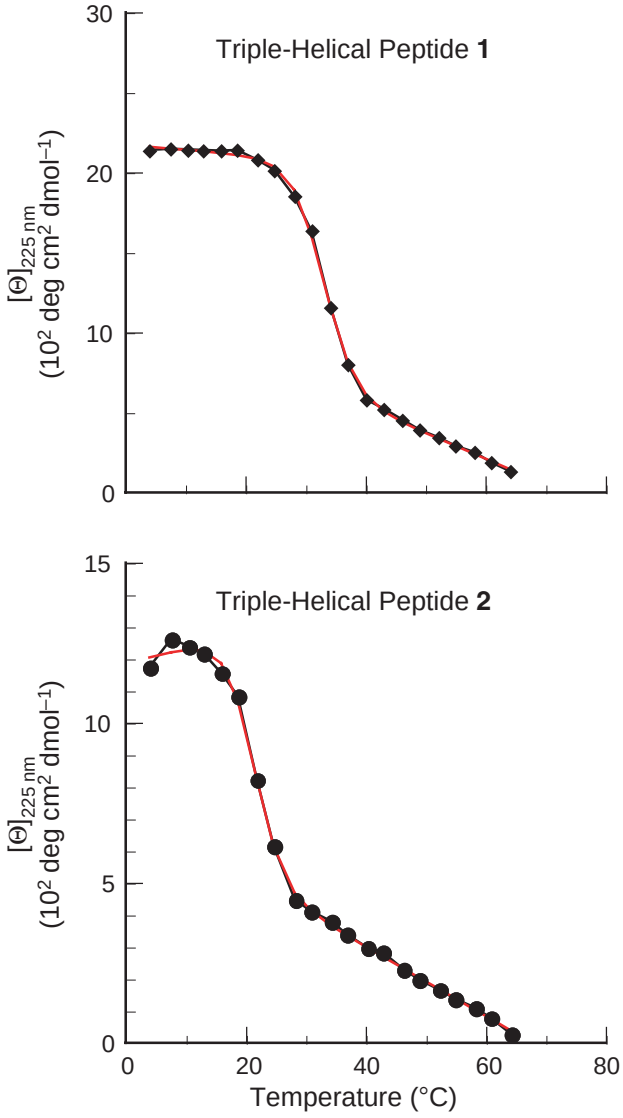

Figure S2. Effect of temperature on the ellipticity at 225 nm of triple helices of peptide **1** and peptide **2**. Red lines are derived from the fit of data to a two-state model for denaturation.

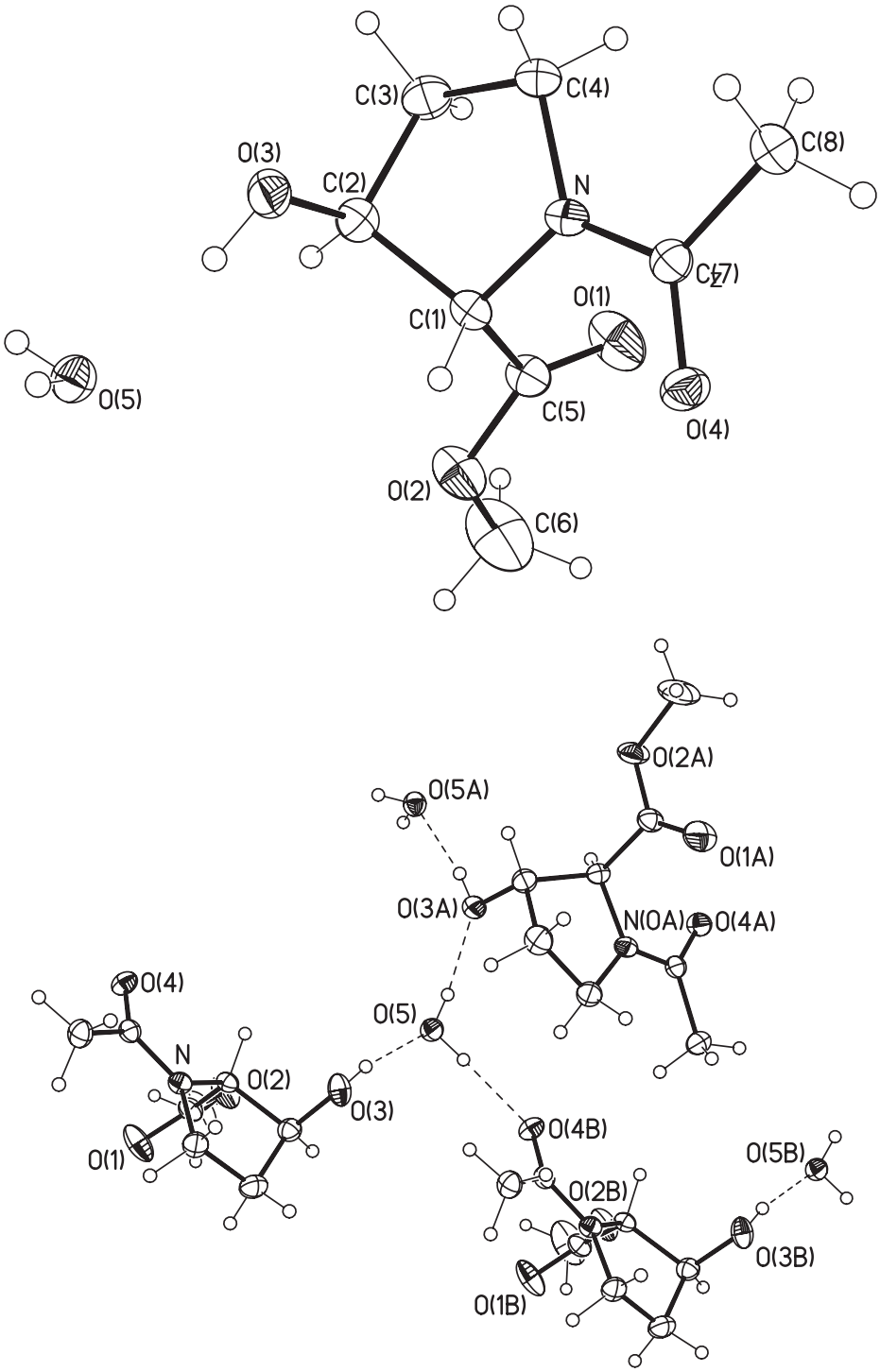

Figure S3. Ortep diagrams of crystalline N-($^{13}C_2$-acetyl)-3(*S*)-hydroxy-L-proline methyl ester (**8**) drawn with 30% probability ellipsoids and atom designations used in Tables S3–S9. Top, asymmetric unit. Bottom, hydrogen bonding scheme.

Table S3. Crystal Data and Structure Refinement for Amide **8**

Identification code	rai04
Empirical formula	$C_8H_{13}NO_4 \cdot H_2O$
Formula weight	205.21
Temperature	173(2) K
Wavelength	0.71073 Å
Crystal system	Orthorhombic
Space group	$P2_12_12_1$
Unit cell dimensions	a = 6.1545(6) Å α = 90°
	b = 8.3050(9) Å β = 90°
	c = 20.807(2) Å γ = 90°
Volume	1063.53(19) Å^3
Z	4
Density (calculated)	1.282 g/cm^3
Absorption coefficient	0.107 mm^{-1}
F(000)	440
Crystal size	0.50 × 0.50 × 0.41 mm^3
Theta range for data collection	1.96–26.37°
Index ranges	$-6 \le h \le 7$, $-9 \le k \le 10$, $-8 \le l \le 26$
Reflections collected	2911
Independent reflections	1963 [R(int) = 0.0126]
Completeness to theta = 26.37°	96.7%
Absorption correction	Empirical with SADABS
Max. and min. transmission	0.9576 and 0.9486
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	1963 / 2 / 138
Goodness-of-fit on F^2	1.051
Final R indices [$I > 2\sigma(I)$]	$R1$ = 0.0369, $wR2$ = 0.0953
R indices (all data)	$R1$ = 0.0401, $wR2$ = 0.0975
Absolute structure parameter	N/A – assigned from synthesis
Largest diff. peak and hole	0.172 and –0.146 e.Å^{-3}

Table S4. Atomic Coordinates ($\times 10^4$) and Equivalent Isotropic Displacement Parameters ($Å^2 \times 10^3$) for Amide **8**

atom	*x*	*y*	*z*	*U*(eq)
O(1)	929(3)	6789(2)	7647(1)	65(1)
O(2)	3794(3)	5154(2)	7753(1)	52(1)
O(3)	838(2)	3244(2)	9362(1)	41(1)
O(4)	2745(2)	8452(2)	9097(1)	36(1)
O(5)	4355(2)	1427(2)	9393(1)	35(1)
N	146(2)	6601(2)	8963(1)	29(1)
C(1)	1640(3)	5476(2)	8661(1)	27(1)
C(2)	462(3)	3857(2)	8736(1)	34(1)
C(3)	–1899(3)	4333(3)	8677(1)	43(1)
C(4)	–2055(3)	5935(2)	9032(1)	38(1)
C(5)	2043(3)	5915(2)	7961(1)	35(1)
C(6)	4383(6)	5434(5)	7081(1)	92(1)
C(7)	826(3)	8039(2)	9168(1)	29(1)
C(8)	–822(3)	9093(2)	9496(1)	39(1)

U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

Table S5. Bond Lengths [Å] and Angles [°] for amide **8**

O(1)–C(5)	1.193(2)
O(2)–C(5)	1.322(2)
O(2)–C(6)	1.463(2)
O(3)–C(2)	1.418(2)
O(4)–C(7)	1.239(2)
N–C(7)	1.336(2)
N–C(1)	1.454(2)
N–C(4)	1.470(2)
C(1)–C(5)	1.522(2)
C(1)–C(2)	1.536(2)
C(2)–C(3)	1.512(3)
C(3)–C(4)	1.524(3)
C(7)–C(8)	1.503(2)
C(5)–O(2)–C(6)	116.03(18)
C(7)–N–C(1)	120.97(15)
C(7)–N–C(4)	126.45(15)
C(1)–N–C(4)	112.53(14)
N–C(1)–C(5)	111.31(14)
N–C(1)–C(2)	102.71(14)
C(5)–C(1)–C(2)	112.61(14)
O(3)–C(2)–C(3)	109.02(16)
O(3)–C(2)–C(1)	109.36(14)
C(3)–C(2)–C(1)	102.49(14)
C(2)–C(3)–C(4)	104.44(16)
N–C(4)–C(3)	102.91(15)
O(1)–C(5)–O(2)	125.43(17)
O(1)–C(5)–C(1)	125.17(18)
O(2)–C(5)–C(1)	109.39(15)
O(4)–C(7)–N	120.56(16)
O(4)–C(7)–C(8)	122.43(16)v
N–C(7)–C(8)	117.00(16)

Symmetry transformations were used to generate equivalent atoms.

Table S6. Anisotropic Displacement Parameters ($Å^2 \times 10^3$) for Amide **8**

atom	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
O(1)	69(1)	87(1)	37(1)	24(1)	3(1)	32(1)
O(2)	58(1)	71(1)	28(1)	8(1)	13(1)	24(1)
O(3)	48(1)	39(1)	37(1)	9(1)	13(1)	11(1)
O(4)	31(1)	34(1)	43(1)	-6(1)	3(1)	-7(1)
O(5)	38(1)	33(1)	34(1)	0(1)	-2(1)	-2(1)
N	23(1)	34(1)	29(1)	0(1)	2(1)	0(1)
C(1)	26(1)	32(1)	23(1)	-2(1)	-2(1)	3(1)
C(2)	35(1)	33(1)	32(1)	-3(1)	0(1)	0(1)
C(3)	33(1)	44(1)	53(1)	-2(1)	-4(1)	-8(1)
C(4)	25(1)	37(1)	52(1)	3(1)	3(1)	-1(1)
C(5)	34(1)	44(1)	28(1)	3(1)	1(1)	4(1)
C(6)	116(3)	127(3)	34(1)	15(1)	33(2)	51(2)
C(7)	31(1)	30(1)	25(1)	3(1)	0(1)	1(1)
C(8)	38(1)	34(1)	43(1)	-3(1)	4(1)	5(1)

The anisotropic displacement factor exponent takes the form: $-2\pi^2[h^2\, a^{*2}U^{11} + \ldots + 2\, h\, k\, a^*\, b^*\, U^{12}]$.

***Table S7*.** Hydrogen Coordinates (x 10^4) and Isotropic Displacement Parameters ($Å^2 \times 10^3$) for Amide **8**

atom	*x*	*y*	*z*	*U*(eq)
H(3)	2046	2774	9372	62
H(5A)	4790(40)	1450(30)	9776(4)	59(7)
H(5B)	3860(30)	505(11)	9315(10)	40(6)
H(1)	3046	5455	8901	32
H(2)	900	3071	8396	40
H(3A)	–2853	3516	8878	52
H(3B)	–2318	4459	8220	52
H(4A)	–3150	6649	8831	45
H(4B)	–2434	5773	9489	45
H(6A)	4680	6582	7016	138
H(6B)	3180	5102	6802	138
H(6C)	5683	4807	6975	138
H(8A)	–217	10174	9554	58
H(8B)	–1188	8634	9916	58
H(8C)	–2136	9156	9231	58

Table S8. Torsion Angles [°] for Amide **8**

C(7)–N–C(1)–C(5)	–79.46(19)
C(4)–N–C(1)–C(5)	103.01(17)
C(7)–N–C(1)–C(2)	159.82(14)
C(4)–N–C(1)–C(2)	–17.72(17)
N–C(1)–C(2)–O(3)	–81.46(16)
C(5)–C(1)–C(2)–O(3)	158.71(15)
N–C(1)–C(2)–C(3)	34.10(17)
C(5)–C(1)–C(2)–C(3)	–85.72(18)
O(3)–C(2)–C(3)–C(4)	77.24(18)
C(1)–C(2)–C(3)–C(4)	–38.58(18)
C(7)–N–C(4)–C(3)	176.70(16)
C(1)–N–C(4)–C(3)	–5.92(19)
C(2)–C(3)–C(4)–N	27.71(19)
C(6)–O(2)–C(5)–O(1)	0.3(4)
C(6)–O(2)–C(5)–C(1)	179.1(2)
N–C(1)–C(5)–O(1)	–17.4(3)
C(2)–C(1)–C(5)–O(1)	97.3(2)
N–C(1)–C(5)–O(2)	163.74(16)
C(2)–C(1)–C(5)–O(2)	–81.54(19)
C(1)–N–C(7)–O(4)	0.9(2)
C(4)–N–C(7)–O(4)	178.12(16)
C(1)–N–C(7)–C(8)	–177.85(15)
C(4)–N–C(7)–C(8)	–0.7(2)

Symmetry transformations used to generate equivalent atoms: #1 $x + 1/2, -y + 1/2, -z + 2$; #2 $x, y - 1, z$

Table S9. Hydrogen Bonds for Amide **8** (Å and °)

D–H⋯A	d(D–H)	d(H⋯A)	d(D⋯A)	∠(D–H–A)
O(3)–H(3)⋯O(5)	0.84	1.81	2.6391(19)	170(1)
O(5)–H(5A)⋯O(3) #1	0.8400(11)	1.922(4)	2.7585(19)	174(2)
O(5)–H(5B)⋯O(4) #2	0.8401(11)	1.893(2)	2.7326(19)	177(2)

Symmetry transformations used to generate equivalent atoms: #1 $x + 1/2, -y + 1/2, -z + 2$; #2 $x, y - 1, z$