Supporting Information for

Importance of backbone conformation of (–)-ternatin on its fat-accumulation inhibitory activity in 3T3-L1 adipocytes

Kenichiro Shimokawa, Ryoka Miwa, Kaoru Yamada, and Daisuke Uemura*

Department of Chemistry, Graduate School of Science,
and Institute for Advanced Research, Nagoya University,
Furo-cho, Chikusa, Nagoya 464-8602, Japan

E-mail: uemura@chem3.chem.nagoya-u.ac.jp

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1. 1D- and 2D-NMR spectra of analogues 1–4 in C_6D_6

	Compound **1**			Compound **2**			Compound **4**		
	Chemical shift [ppm]			Chemical shift [ppm]			Chemical shift [ppm]		
	Position		$^3J_{\alpha,NH}$	Position		$^3J_{\alpha,NH}$	Position		$^3J_{\alpha,NH}$
Residue	NH	α	[Hz]	NH	α	[Hz]	NH	α	[Hz]
D-*allo*-Ile[1]	6.22 (H_D, br d)	4.48 (dd)	7.2	6.64 (H_D, br d)	4.92 (dd)	9.5	6.92-6.88 (H_D, br d)	4.57 (t)	5.9
*N*Me-L-Ala[2]	–	5.68 (q)	–	–	5.32 (q)	–	–	5.75 (q)	–
*N*Me-L-Leu[3]	–	4.31 (dd)	–	–	5.58 (dd)	–	–	4.47 (m)	–
L-Leu[4][a]	7.57 (H_A, d)	5.20 (dt)	8.0				7.89 (H_A, d)	5.20 (ddd)	8.0
(*N*Me-L-Leu[4])[b]				–	6.08 (t)	–			
*N*Me-L-Ala[5]	–	4.34 (q)	–	–	5.44 (q)	–	–	4.25 (q)	–
*N*Me-D-Ala[6]	–	5.64 (q)	–	–	5.65 (q)	–	–	5.61 (q)	–
β-OH-D-Leu[7][c]	7.92 (H_B, d)	5.17 (dt)	8.0	6.80 (H_B, d)	4.38 (dd)	4.0			
(D-Leu[7])[d]							7.98 (H_B, d)	5.20-5.10 (m)	8.5
	OH 5.87 (H_C, s)	β 3.96 (dd)		OH 3.23 (H_C, s)	β 3.16-3.13 (m)		–	–	

Table S1. The key ^{1}H NMR parameter for compounds **1**, **2**, and **4** in C_6D_6.
[a]amino acid residue for **1** and **4**. [b]Amino acid residue for **2**. [c]Amino acid residue for **1** and **2**. [d]Amino acid residue for **4.**

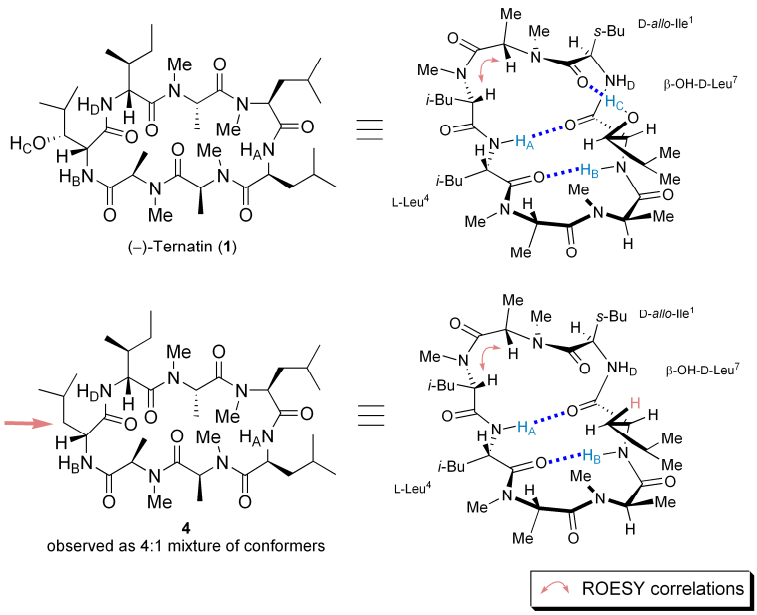


Figure S1. The key ROESY correlations (measured in C_6D_6) for compounds **1** and **4** that indicated the existence of unique cis-amide bond regions. The ROESY spectra were available in ref 6c and SI therein for **1**, and figureS1-4-3 for **4**.

1-1. NMR spectra of (–)-ternatin (1)

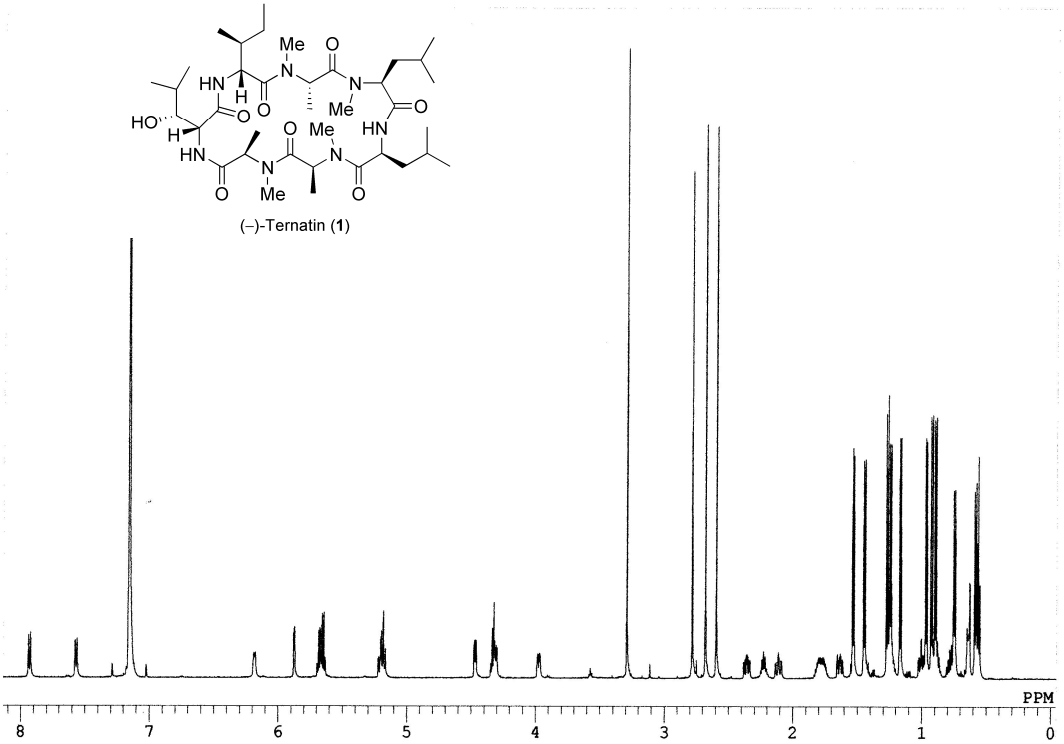


Figure S1-1-1. ^{1}H NMR spectrum of (–)-ternatin (**1**) in C_6D_6 (600 MHz)

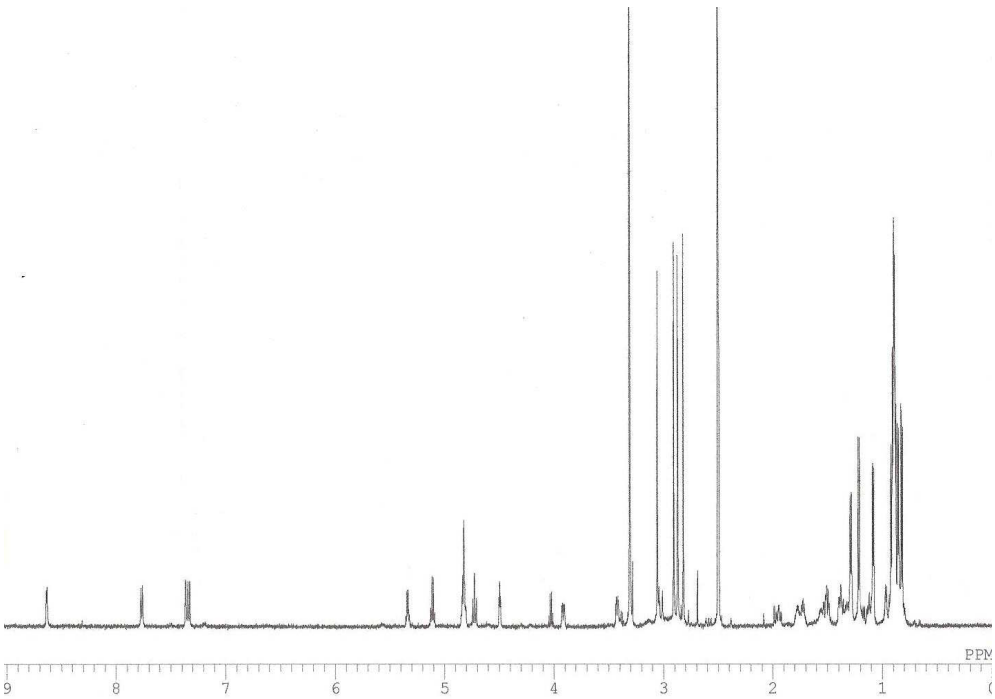


Figure S1-1-2. ^{1}H NMR spectrum of (–)-ternatin (**1**) in DMSO-d_6 (600 MHz)

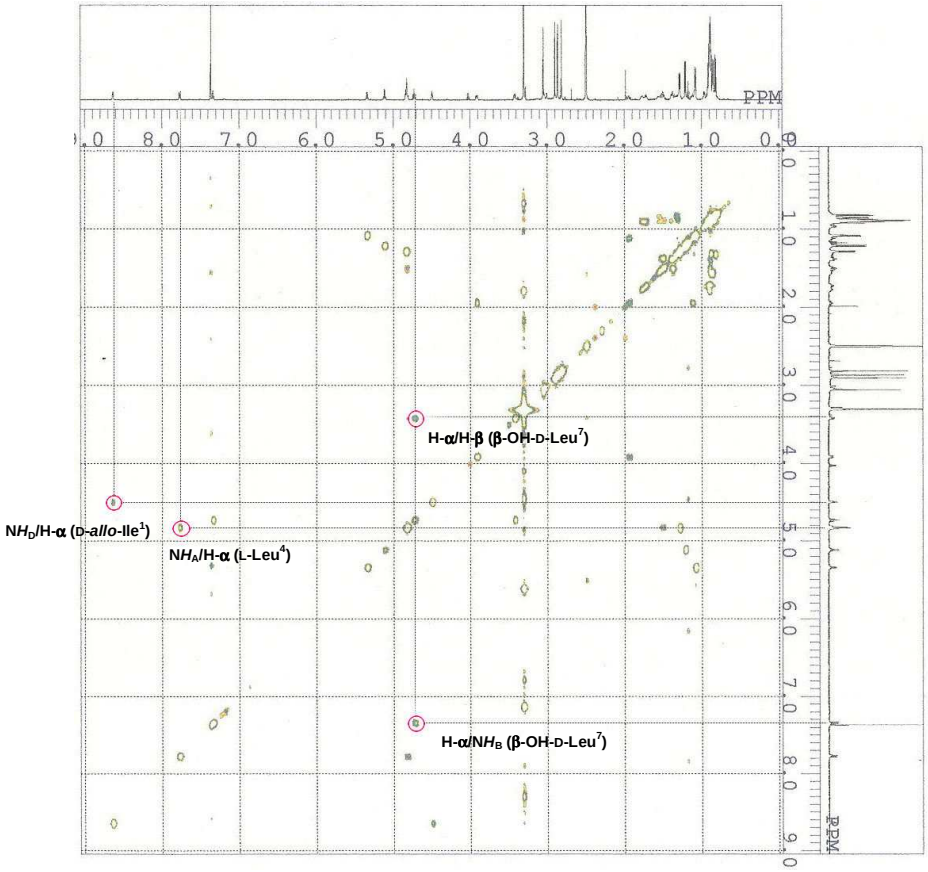


Figure S1-1-3. COSY spectrum of (–)-ternatin (**1**) in DMSO-d_6 (600 MHz)

Residue	Chemical shift [ppm]				
	Position			Extra	
	NH	α	$^3J_{\alpha,NH}$ [Hz]	OH	β
D-*allo*-Ile[1]	8.63 (H_D, br d)	4.49 (dd)	4.7		
L-Leu[4]	7.76 (H_A, d)	4.84-4.79 (dt)	8.8		
β-OH-D-Leu[7]	7.33 (H_B, d)	4.72 (dd)	9.9	4.81 (H_C, d, $^3J_{\beta,OH}$ = 2.2)	3.41 (dd)

Table S1-1-1. Key ^{1}H NMR parameters for (–)-ternatin (**1**) in DMSO-d_6.

1-2. NMR spectra of [*N*Me-L-Leu^4]ternatin (2)

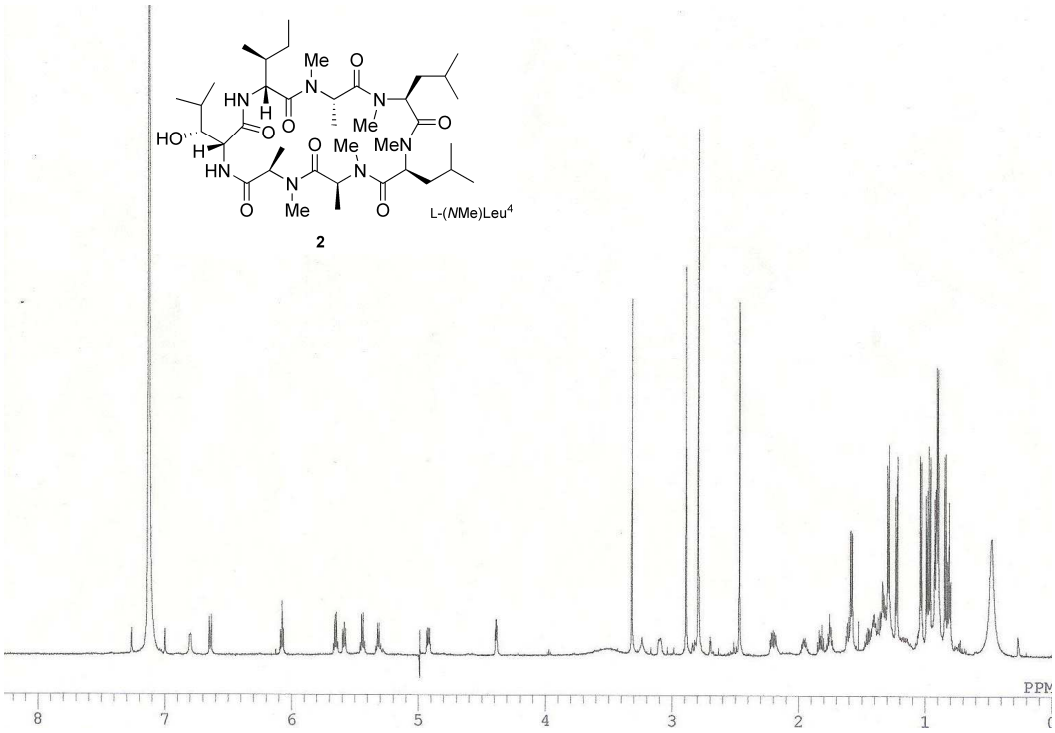


Figure S1-2-1. ^{1}H NMR spectrum of [*N*Me-L-Leu^4]ternatin (**2**) in C_6D_6 (600 MHz)

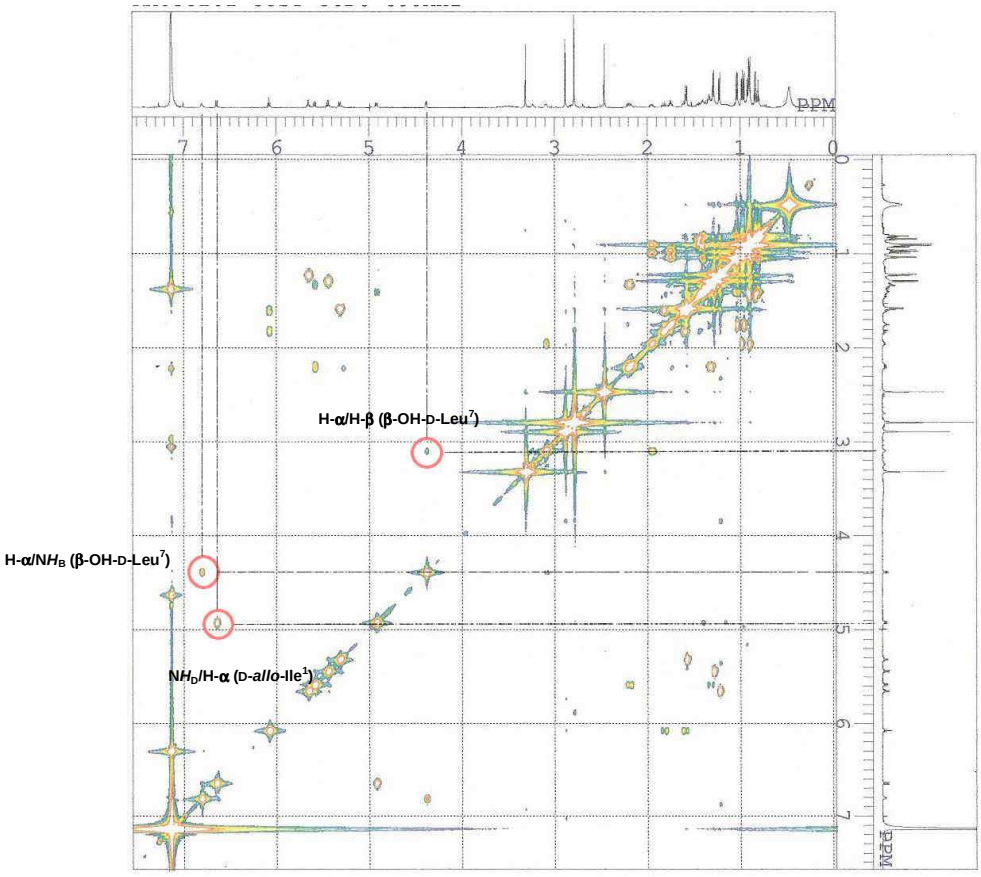


Figure S1-2-2. COSY spectrum of [*N*Me-L-Leu^4]ternatin (**2**) in C_6D_6 (600 MHz)

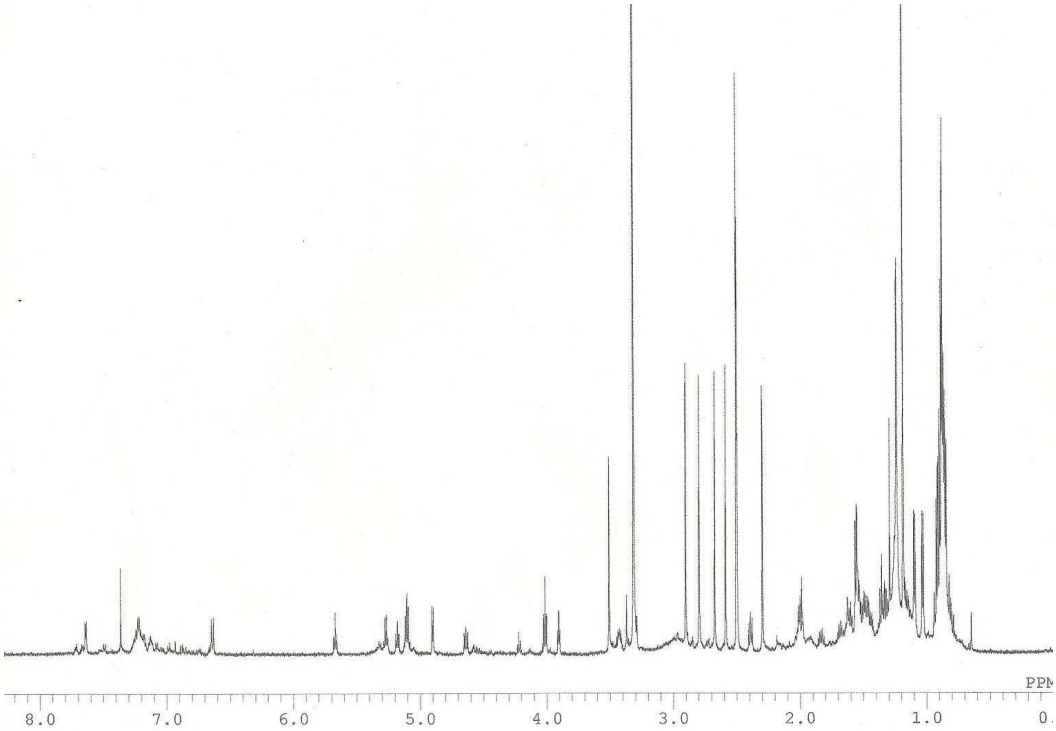


Figure S1-2-3. ^{1}H NMR spectrum of [*N*Me-L-Leu^{4}]ternatin (**2**) in DMSO-d_6 (600 MHz)

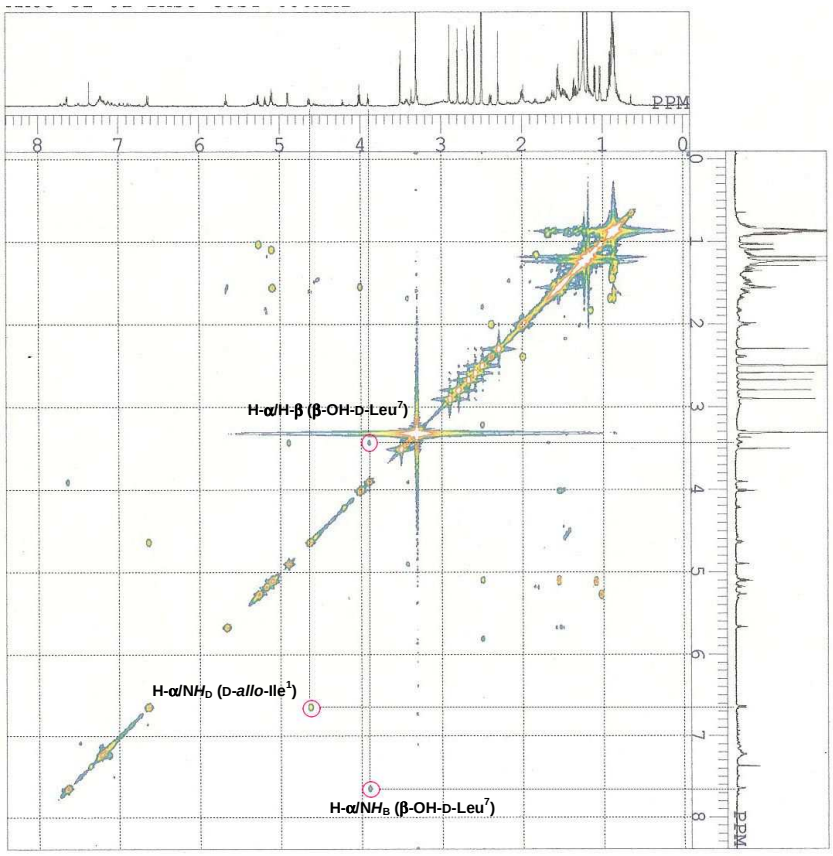


Figure S1-2-4. COSY spectrum of [*N*Me-L-Leu^{4}]ternatin (**2**) in DMSO-d_6 (600 MHz)

Residue	Chemical shift [ppm]				
	Position			Extra	
	NH	α	$^3J_{\alpha,NH}$ [Hz]	OH	β
D-*allo*-Ile[1]	6.64 (H_D, d)	4.64 (dd)	9.2		
β-OH-D-Leu[7]	7.64 (H_B, d)	3.91 (dd)	5.2	4.90 (H_C, d, $^3J_{\beta,OH}$ = 2.2)	3.46-3.40 (m)

Table S1-2-2. Key ^{1}H NMR parameters for [*N*Me-L-Leu[4]]ternatin (**2**) in DMSO-d_6

1-3. NMR spectra of [*N*Me-β-OH-D-Leu[7]]ternatin (3)

Due to the complexity of the spectrum as shown below, further analysis of compound **3** was not available.

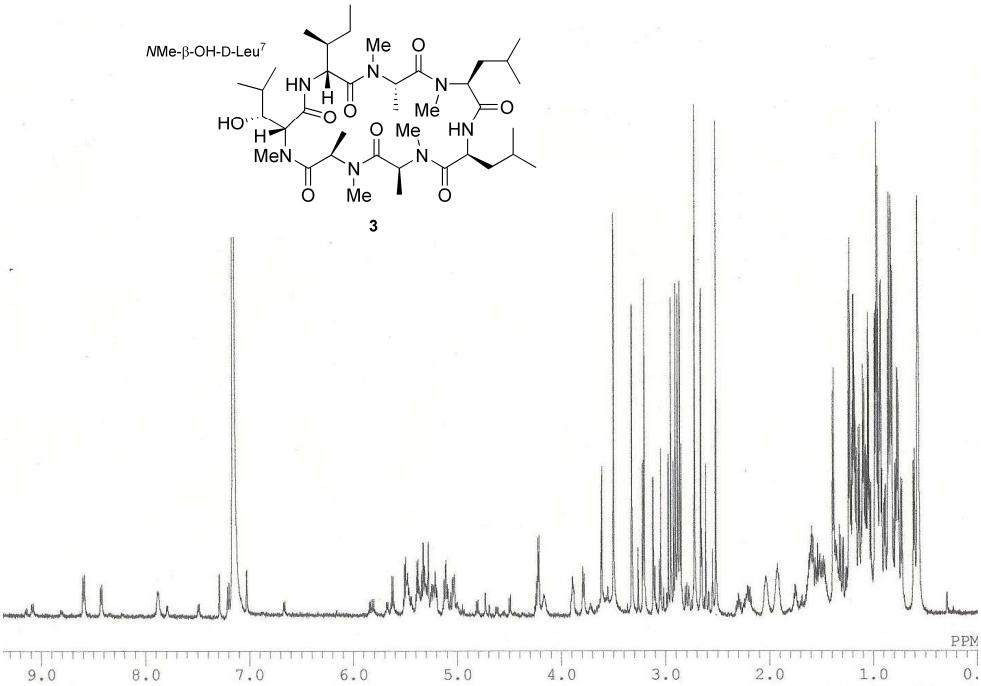


Figure S1-3-1. ^{1}H NMR spectrum of [*N*Me-β-OH-D-Leu[7]]ternatin (**3**) in C_6D_6 (600 MHz)

1-4. NMR spectra of [D-Leu^7]ternatin (4)

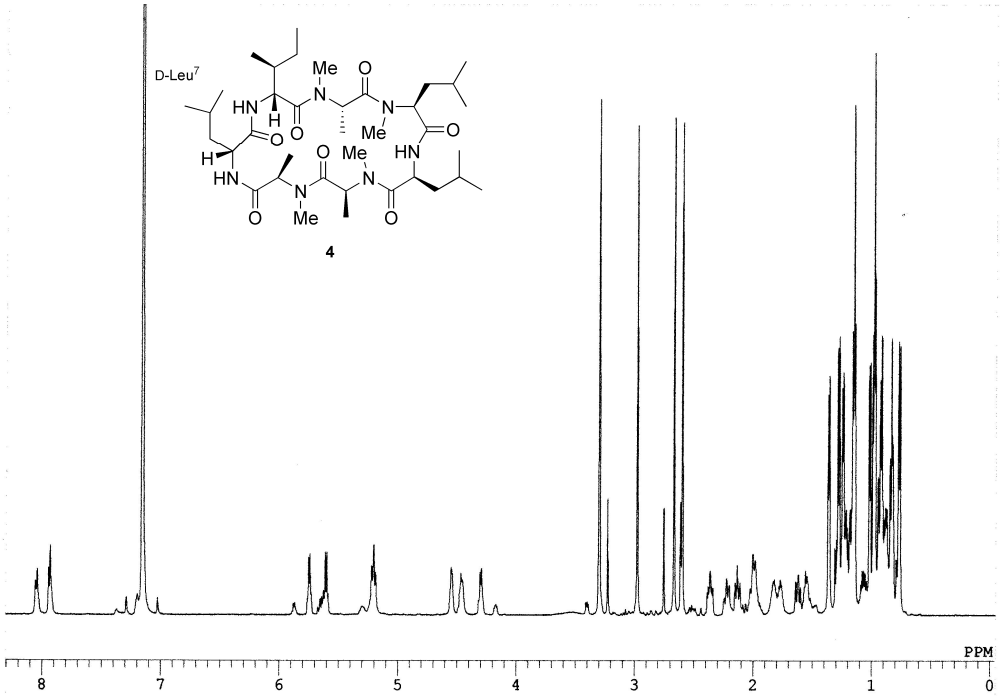


Figure S1-4-1. ^{1}H NMR spectrum of [D-Leu^7]ternatin (**4**) in C_6D_6 (600 MHz)

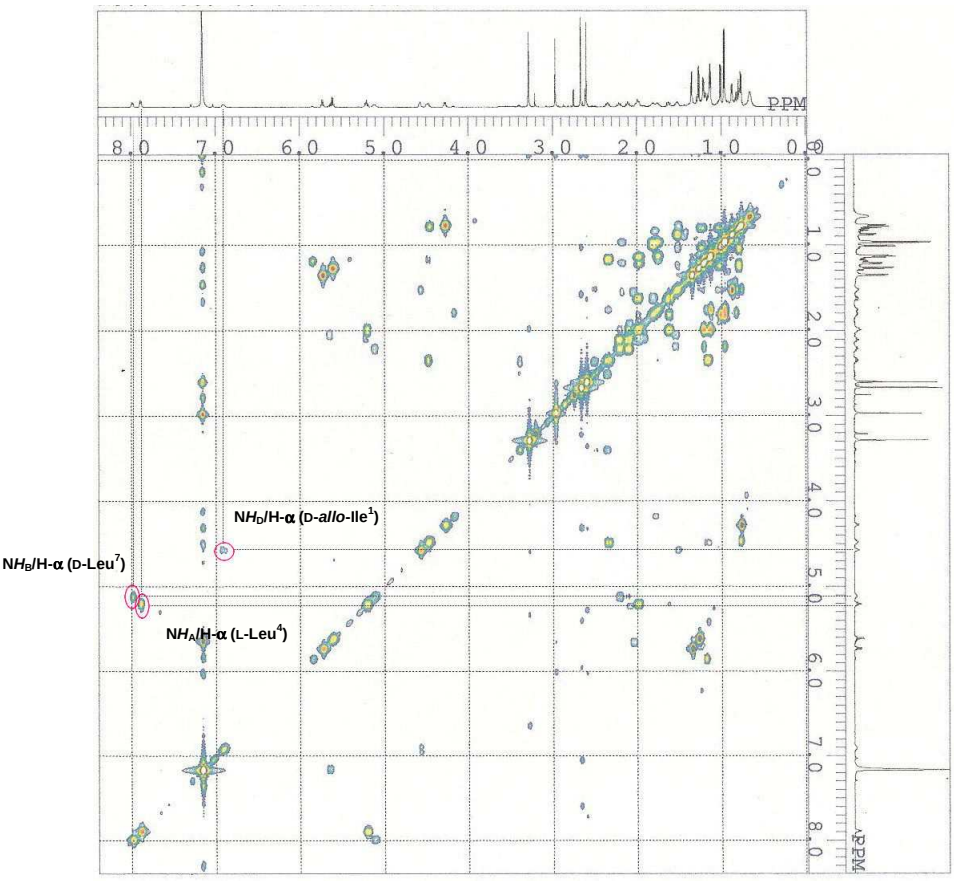


Figure S1-4-2. COSY spectrum of [D-Leu^7]ternatin (**4**) in C_6D_6 (600 MHz)

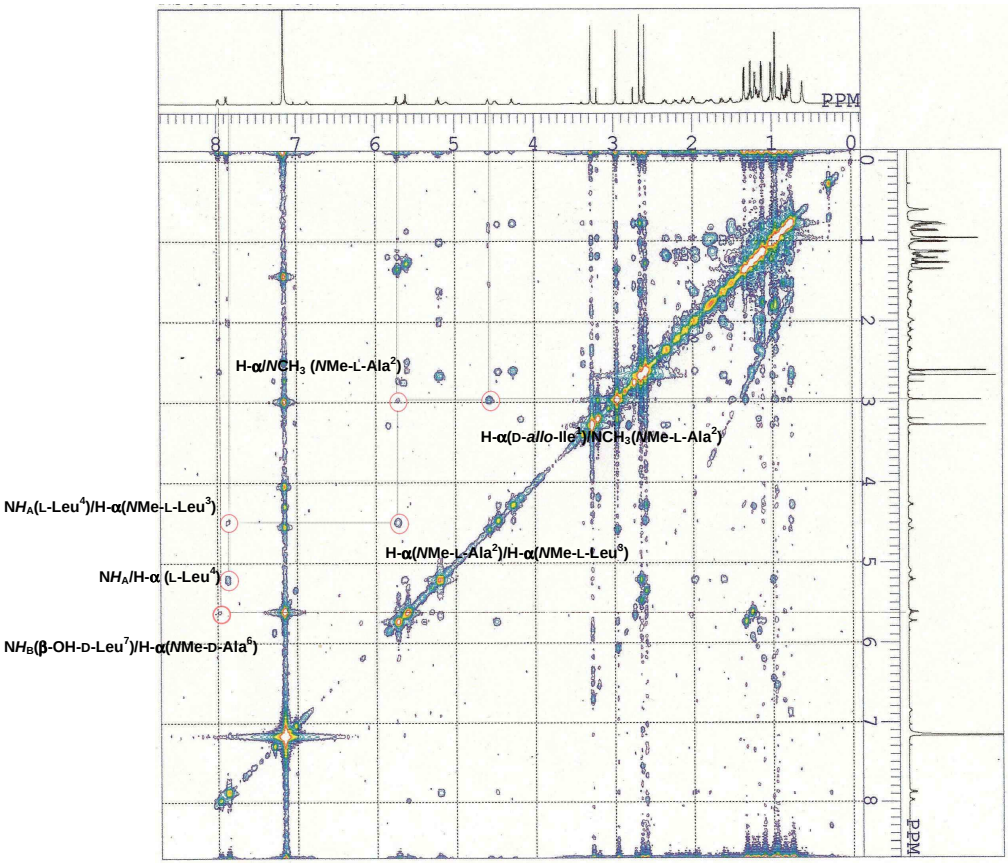


Figure S1-4-3. ROESY spectrum of [D-Leu^7]ternatin (**4**) in C_6D_6 (600 MHz)

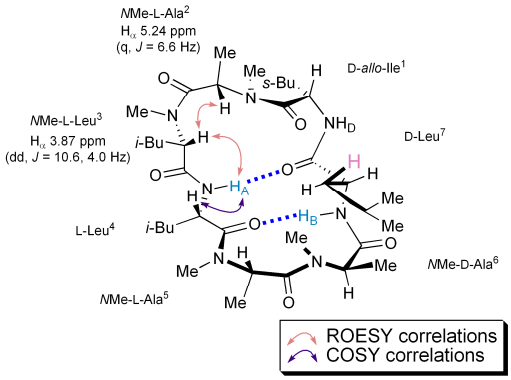


Figure S1-4-4. Key ROESY correlations to distinguish amide protons H_A and H_B of [D-Leu^7]ternatin (**4**) in C_6D_6

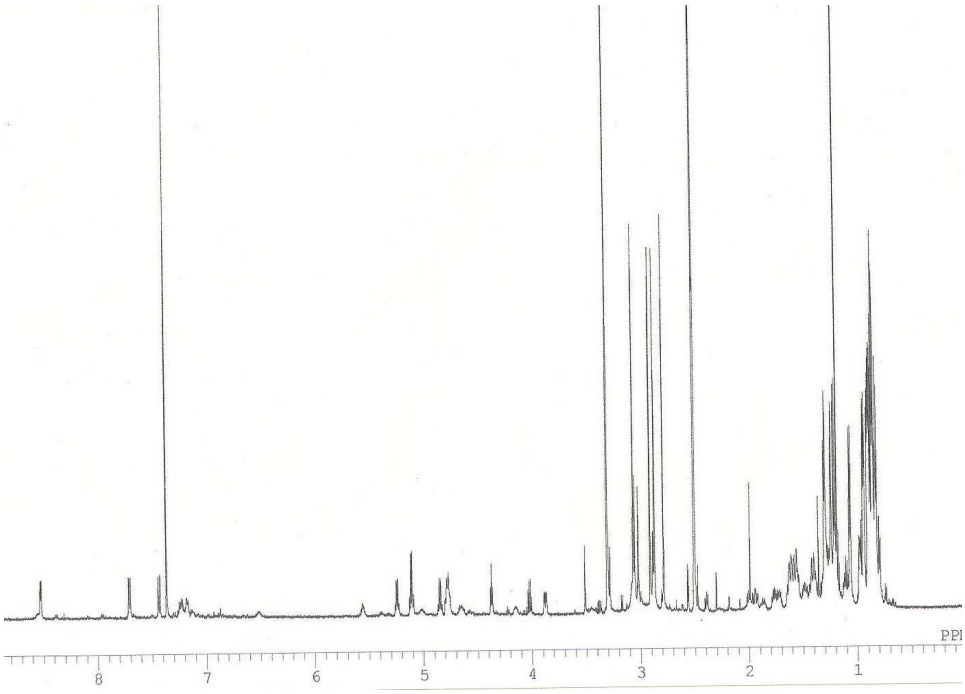


Figure S1-4-5. ^{1}H NMR spectrum of [D-Leu^{7}]ternatin (**4**) in DMSO-d_6 (600 MHz). The minor but multiple conformers existed in **2** in DMSO-d_6, however, major conformer existed as a dominant one.

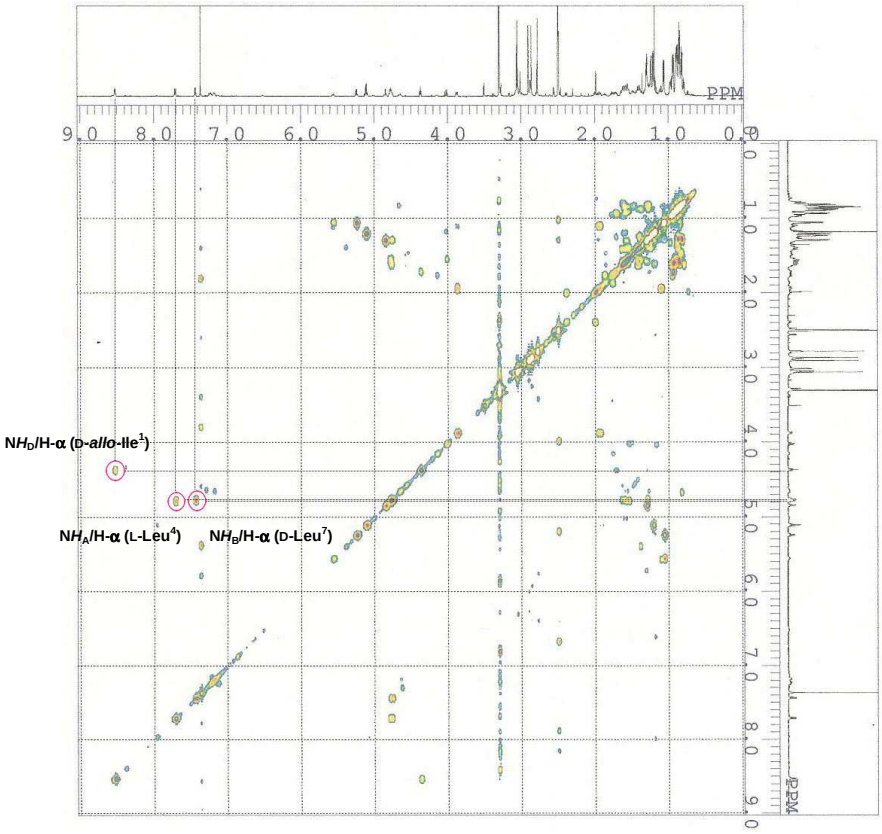


Figure S1-4-6. COSY spectrum of [D-Leu^{7}]ternatin (**4**) in DMSO-d_6 (600 MHz)

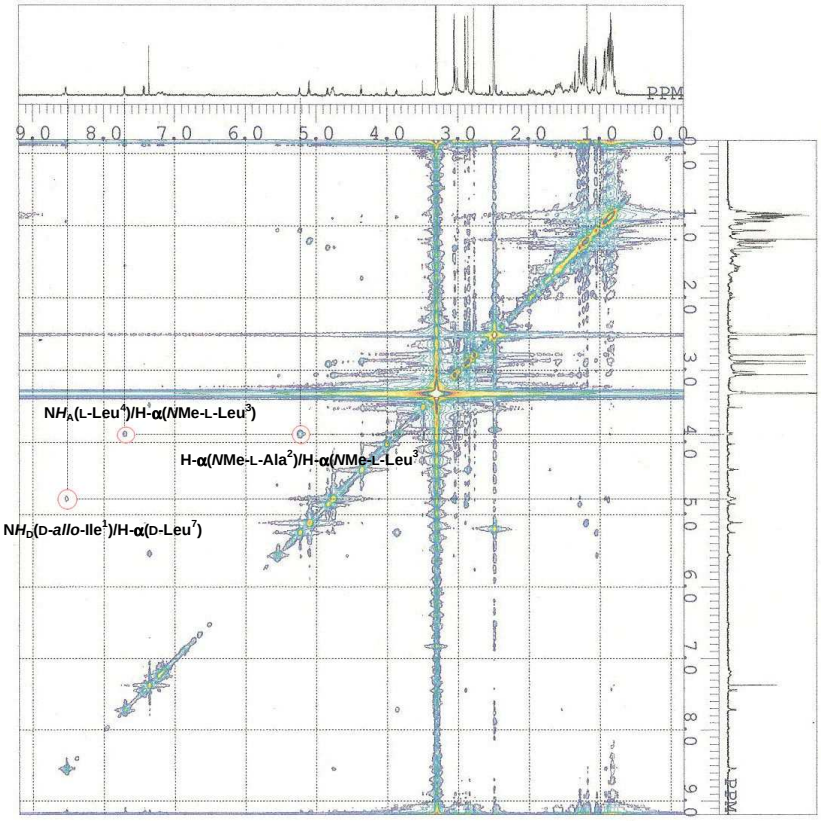


Figure S1-4-7. ROESY spectrum of [D-Leu^7]ternatin (**4**) in DMSO-d_6 (600 MHz)

Residue	Chemical shift [ppm]				
	Position			Extra	
	NH	α	$^3J_{\alpha,NH}$ [Hz]	OH	β
D-*allo*-Ile^1	8.53 (H_D, d)	4.37 (dd)	5.5		
L-Leu^4	7.71 (H_A, d)	4.81-4.75 (m)	8.8		
D-Leu^7	7.43 (H_B, d)	4.81-4.75 (dd)	9.5	–	–

Table S1-4-2. Key ^{1}H NMR parameters for [D-Leu^7]ternatin (**4**) in DMSO-d_6

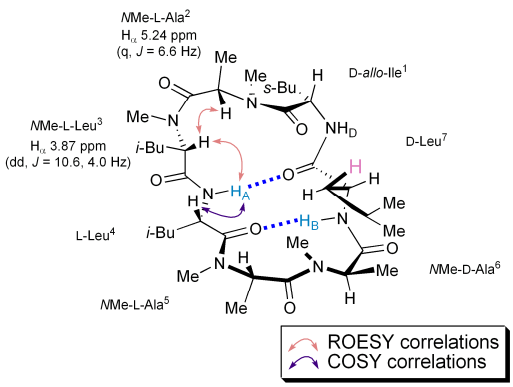


Figure S1-4-8. Key ROESY correlations to distinguish amide protons H_A and H_B of [D-Leu^7]ternatin (**4**) in DMSO-d_6

2. H/D exchange experiment

Deuterium exchange experiment was carried out on a 600 MHz (JNM-A600) at room temperature. Compounds **1**,[5] **2**, and **4** was dissolved in 650 μL of C_6D_6. Before addition of D_2O, a proton spectra was taken (0 min). Next, 20 μL of D_2O was added to the NMR tube and shaken vigorously. Then, proton spectra were taken at several time points (10 min, 1 h, 5 h, 10 h, 24 h).

2-1. H-D-exchange experiment of [*N*Me-L-Leu[4]]ternatin (2)

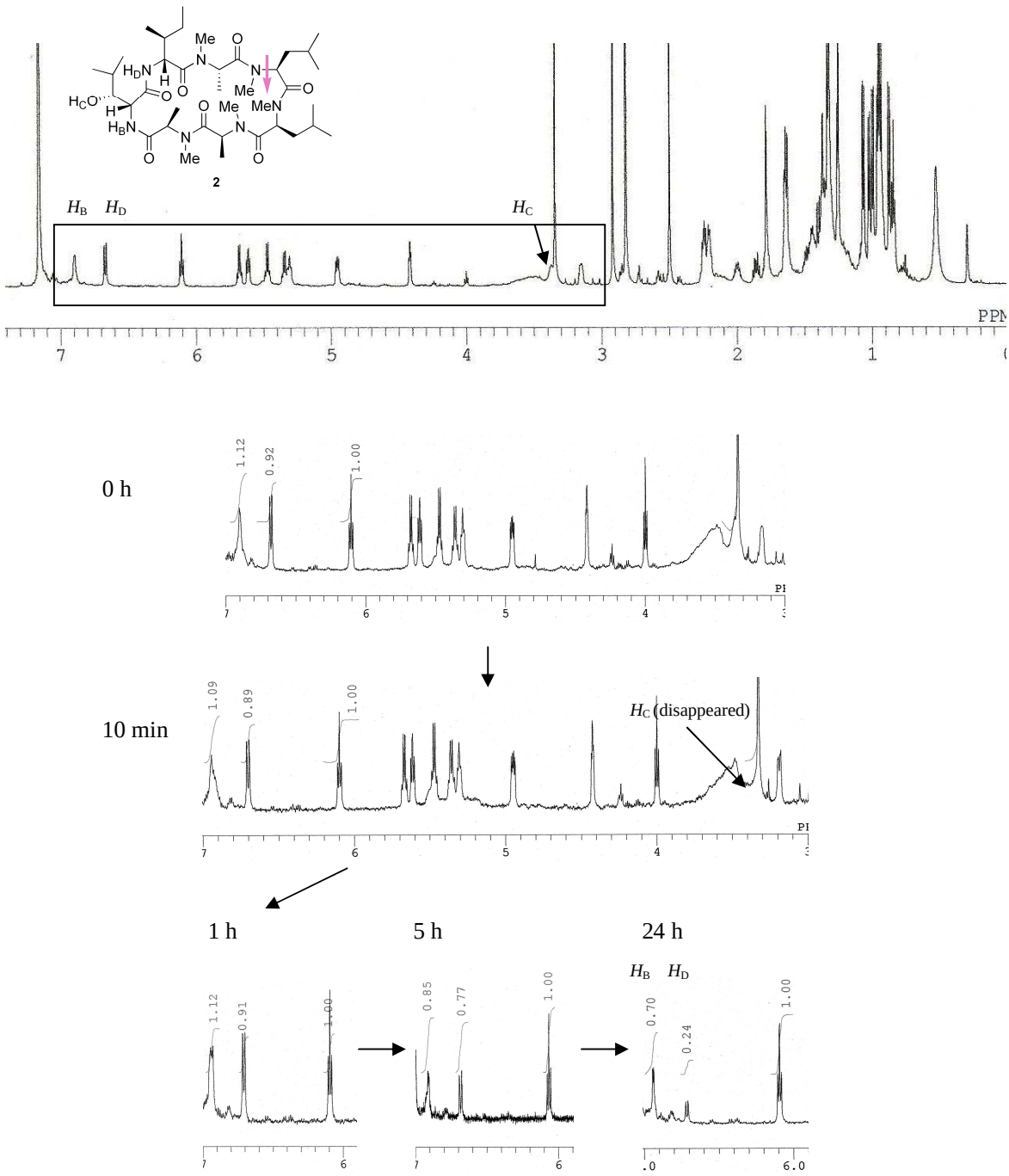
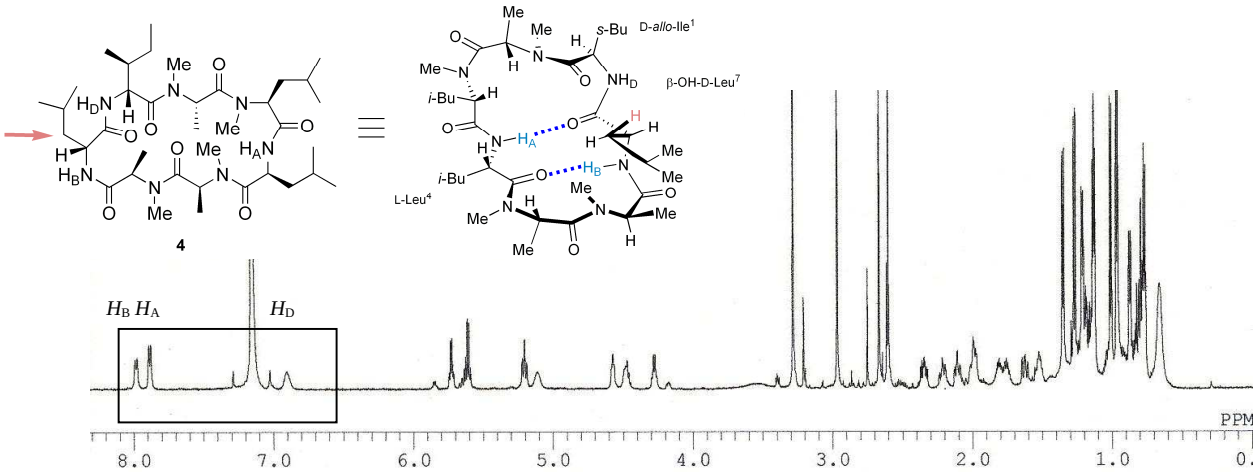


Figure S2-1. H–D exchange experiment of [*N*Me-L-Leu[4]]ternatin (**2**) in C_6D_6 (600 MHz)

2-2. H-D-exchange experiment of [D-Leu^7]ternatin (4)



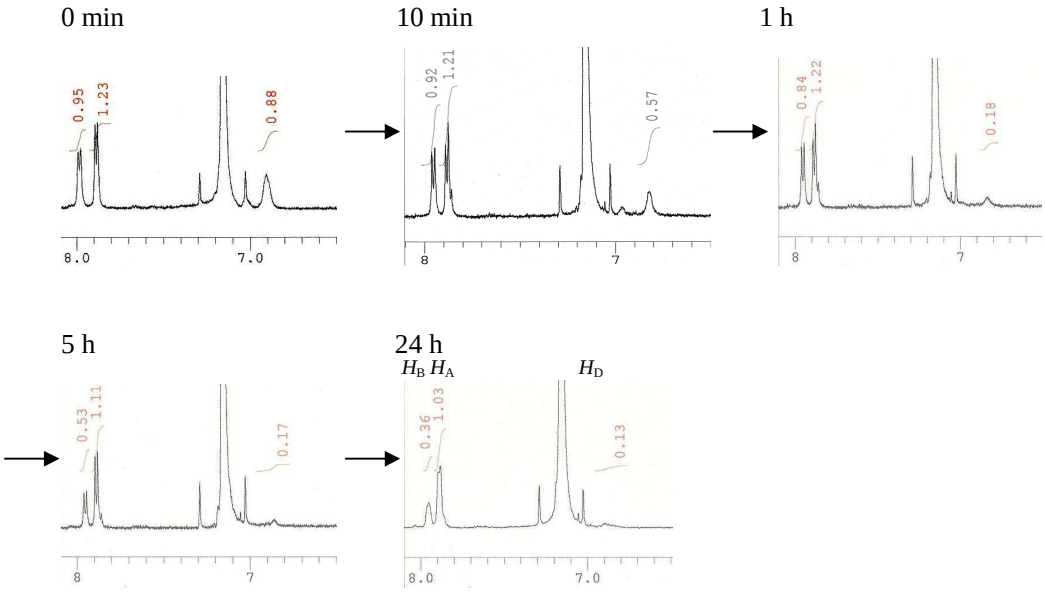


Figure S2-2. H–D exchange experiment of [D-Leu^7]ternatin (**4**) in C_6D_6 (600 MHz)

3. Measurement of temperature coefficient of amide protons

The experiment was carried out on a 600 MHz (JNM-A600). ^{1}H NMR spectra of all compounds **1**, **2**, and **4** were taken in DMSO-d_6 at several temperatures (25, 30, 40, 50, 60 °C).

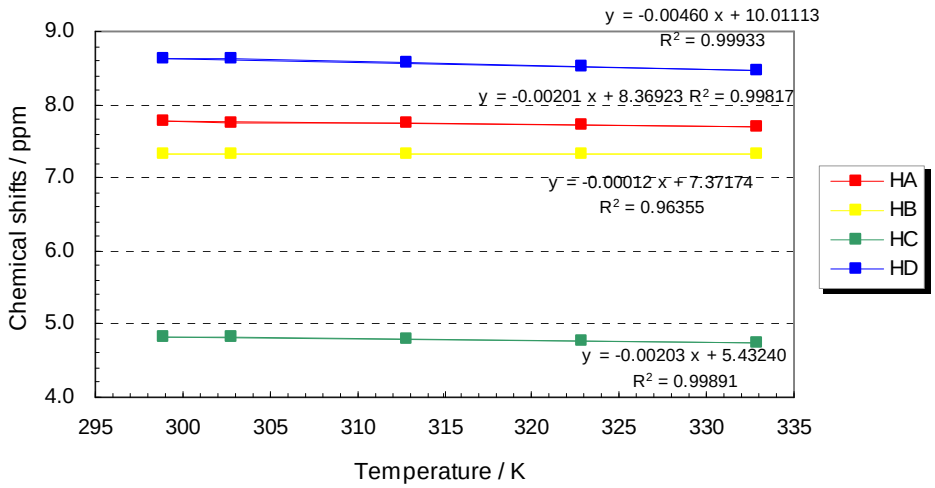


Figure S3-1. Chemical shifts of NH protons (H_A, H_B, and H_D) and a OH proton (H_C), of (–)-ternatin (**1**) in the range of 295-335 K

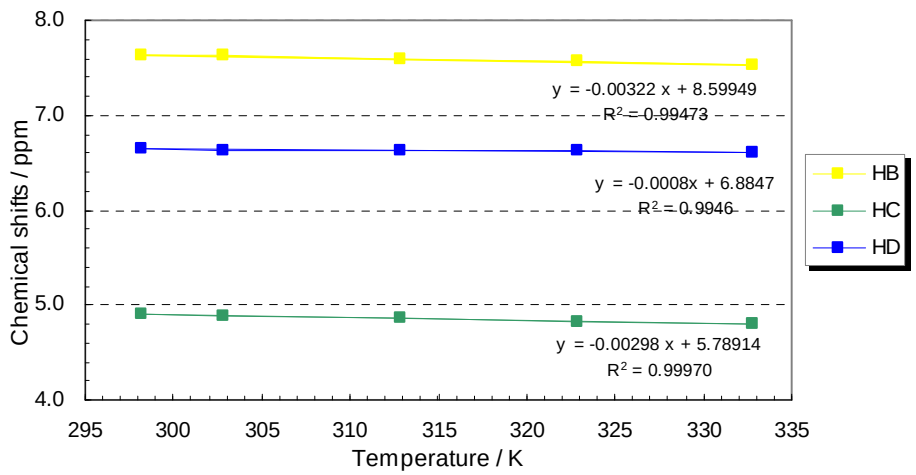


Figure S3-2. Chemical shifts of NH protons (H_B and H_D) and a OH proton (H_C) of [*N*Me-L-Leu^4]ternatin (**2**) in the range of 295-335 K

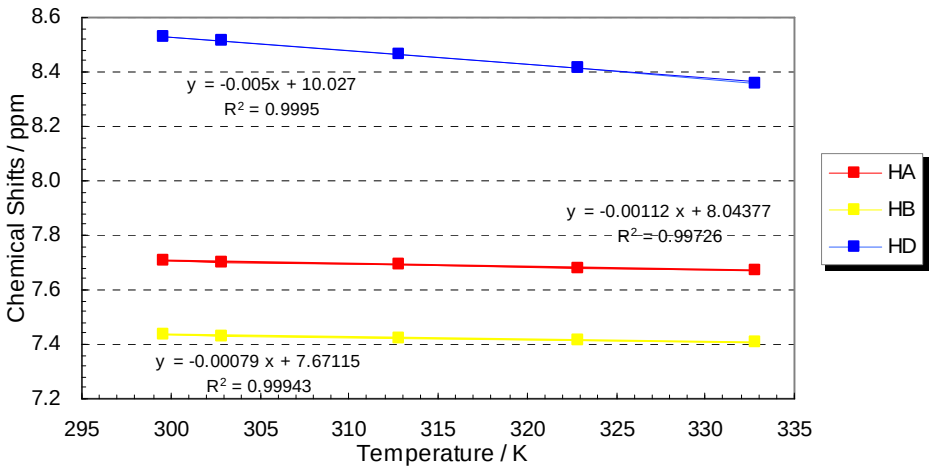


Figure S3-3. Chemical shifts of NH protons (H_A, H_B, and H_D) of [D-Leu^7]ternatin (**4**) in the range of 295-335 K

4. CD spectra

CD measurements were obtained on a JASCO J-720 spectrometer. For these experiments, each sample was dissolved in MeOH (c = 0.1 mg/mL, 0.1 cm path length, 25 °C).