Supplementary Materials

Characterization of the ohmyungsamycin biosynthetic pathway and generation of the derivatives with improved antituberculosis activity

Eunji Kim [1,†], Yern-Hyerk Shin [2,†], Tae Ho Kim [3], Woong Sub Byun [2], Jinsheng Cui [2], Young Eun Du [2], Hyung-Ju Lim [2], Myoung Chong Song [1], An Sung Kwon [4], Sang Hyeon Kang [4], Jongheon Shin [2], Sang Kook Lee [2], Jichan Jang [3], Dong-Chan Oh [2,*] and Yeo Joon Yoon [1,*]

[1] Department of Chemistry and Nanoscience, Ewha Womans University, Seoul 03760, Republic of Korea; ejkim0618@ewha.ac.kr (E.K.); smch517@ewha.ac.kr (M.C.S.)

[2] Natural Products Research Institute, College of Pharmacy, Seoul National University, Seoul 08826, Republic of Korea; itsue00@snu.ac.kr (Y.-H.S.); sky_magic@naver.com (W.S.B.); cuijs@snu.ac.kr (J.C.); dye0302@snu.ac.kr (Y.E.D.); limju012@snu.ac.kr (H.-J.L.); shinj@snu.ac.kr (J.S.); sklee61@snu.ac.kr (S.K.L.)

[3] Division of Applied Life Science (BK21plus Program), Gyeongsang National University, Jinju 52828, Republic of Korea; taeho12349@gmail.com (T.H.K.); jichanjang@gnu.ac.kr (J.J.)

[4] iNtRON Biotechnology, Inc., Seongnam-si, Gyeonggi-do 13202, Republic of Korea; kwon4053@intron.co.kr (A.S.K.); kangsh0403@naver.com (S.H.K)

* Correspondence: dongchanoh@snu.ac.kr; Tel.: +82-2-880-2491 (D.-C.O.)
joonyoon@ewha.ac.kr; Tel.: +82-2-3277-4082 (Y.J.Y.)

† These authors contributed equally.

Table of Contents

Figure S1. Conserved sequence regions from alignment comparisons of P450s implicated in the *β*-hydroxylation of PCP-bound amino acid residues. Numbering is indicated for OhmL. Identity residues are shown as bold and blue. Similar residues and exceptions are shown in normal and italics, respectively.

P450s	B-B_2 loop N-term.		B-B_2 loop C-term.						F-helix				G-helix					I-helix			β-1 sheet		
	69	72	82	83	84	86	89	90	169	170	171	172	185	186	187	188	191	230	231	237	284	285	286
OhmL	**G**	L	A	A	**G**	**M**	V	T	T	**A**	F	G	**A**	H	*V*	*D*	S	**N**	**C**	**G**	A	M	**H**
Ecu1	**G**	L	A	A	*D*	**M**	V	T	V	**A**	F	G	**A**	H	*V*	**E**	S	**N**	**C**	**G**	A	M	**H**
Sky32	**G**	L	A	A	**G**	**M**	V	T	S	**A**	L	S	**A**	R	N	**E**	L	**N**	**C**	**G**	A	M	**H**
OxyD	**G**	I	S	G	**G**	**M**	V	S	H	**A**	F	G	**A**	H	T	**E**	V	**N**	**C**	**G**	A	M	**H**
NovI	**G**	L	A	S	**G**	**M**	V	T	H	**A**	W	S	**A**	K	N	**E**	L	**N**	**C**	**G**	S	L	**H**
NikQ	**G**	L	A	A	**G**	**M**	I	T	F	**A**	W	S	**A**	H	T	**E**	L	**N**	**C**	**G**	V	M	**H**
Ecm12	**G**	L	A	G	**G**	**M**	V	T	L	**A**	L	S	**A**	R	N	**E**	G	**N**	**C**	**G**	A	M	**H**
Consensus	**G**	I L	S G A	S G A	**G**	**M**	V I L	S T	H	**A**	W F L	S G A	**A**	K R H	N T S	**E**	L V S G	**N**	**C**	**G**	S A V	L M	**H**

Accession numbers: Ecu1 (AIW58896.1, *Nonomuraea* sp. MJM5123), Sky32 (AEA30275.1, *Streptomyces* sp. Acta 2897), OxyD (CAC48370.1, *Amycolatopsis balhimycina* DSM 5908), NovI (AAF67502.1, *Streptomyces niveus* NCIMB 9219), NikQ (CAB75339.1, *Streptomyces tendae* Tue901), Ecm12 (BAE98161.1, *Streptomyces lasaliensis*)

Figure S2A. Structures of marine-derived cyclic peptides containing 5-hydroxy-L-Trp moiety. Hydroxy groups at position 5 on the indole are marked in blue.

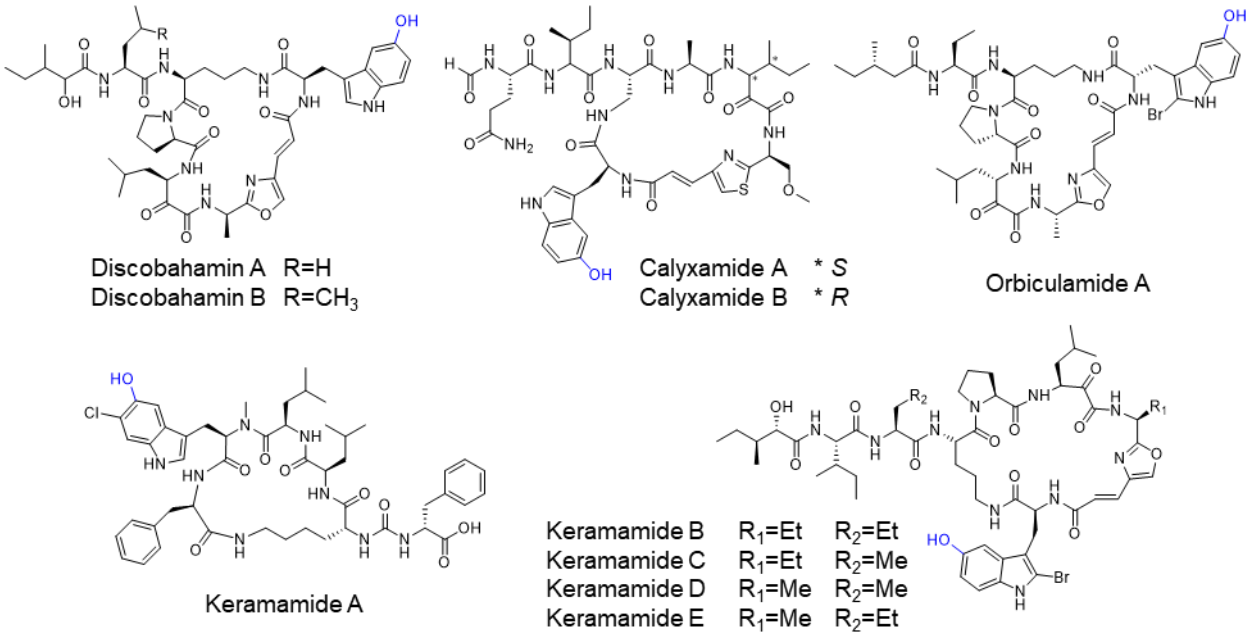


Figure S2B. Structures of compounds containing L-Trp derivatives which are generated by Trp 2,3-dioxygenase catalysis via the kynurenine pathway. Disguised Trp moieties are indicated in red.

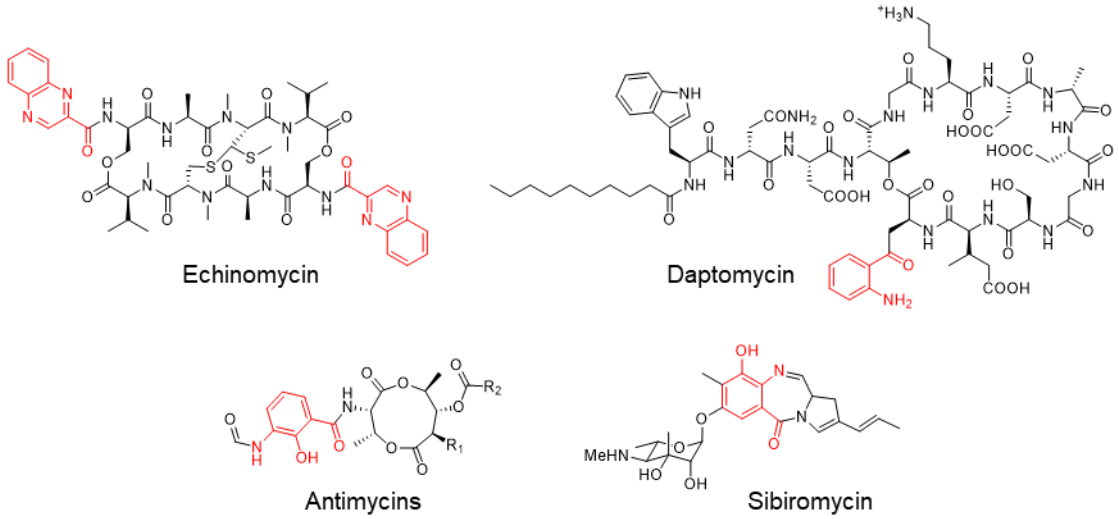


Figure S3. Schematic representations of mutant strain construction by in-frame deletion.

Construction of modification gene deletion mutant strains *ΔohmL*(i), *ΔohmK* (iii), and *ΔohmJ* (v), respectively, and verification of each mutant strain by PCR analysis (ii), (iv), and (vi).

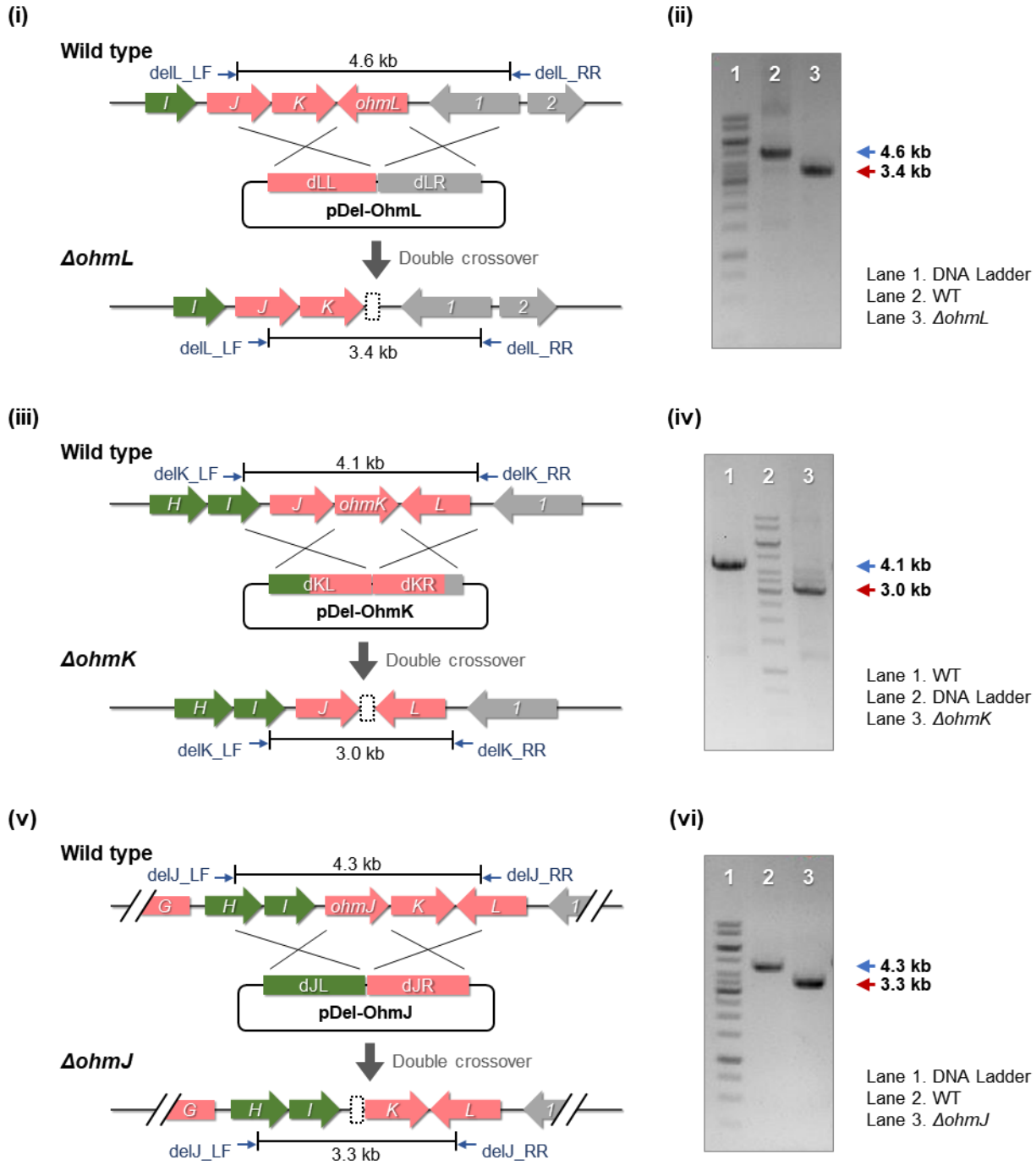


Figure S4A. Structural assignment of OMS A (**1**) and OMS B (**2**) produced from SNJ042 wild type.

(i) MS/MS fragmentation pattern and MS/MS spectra of OMS A (**1**) from wild type strain.
(ii) MS/MS fragmentation pattern and MS/MS spectra of OMS B (**2**) from wild type strain.

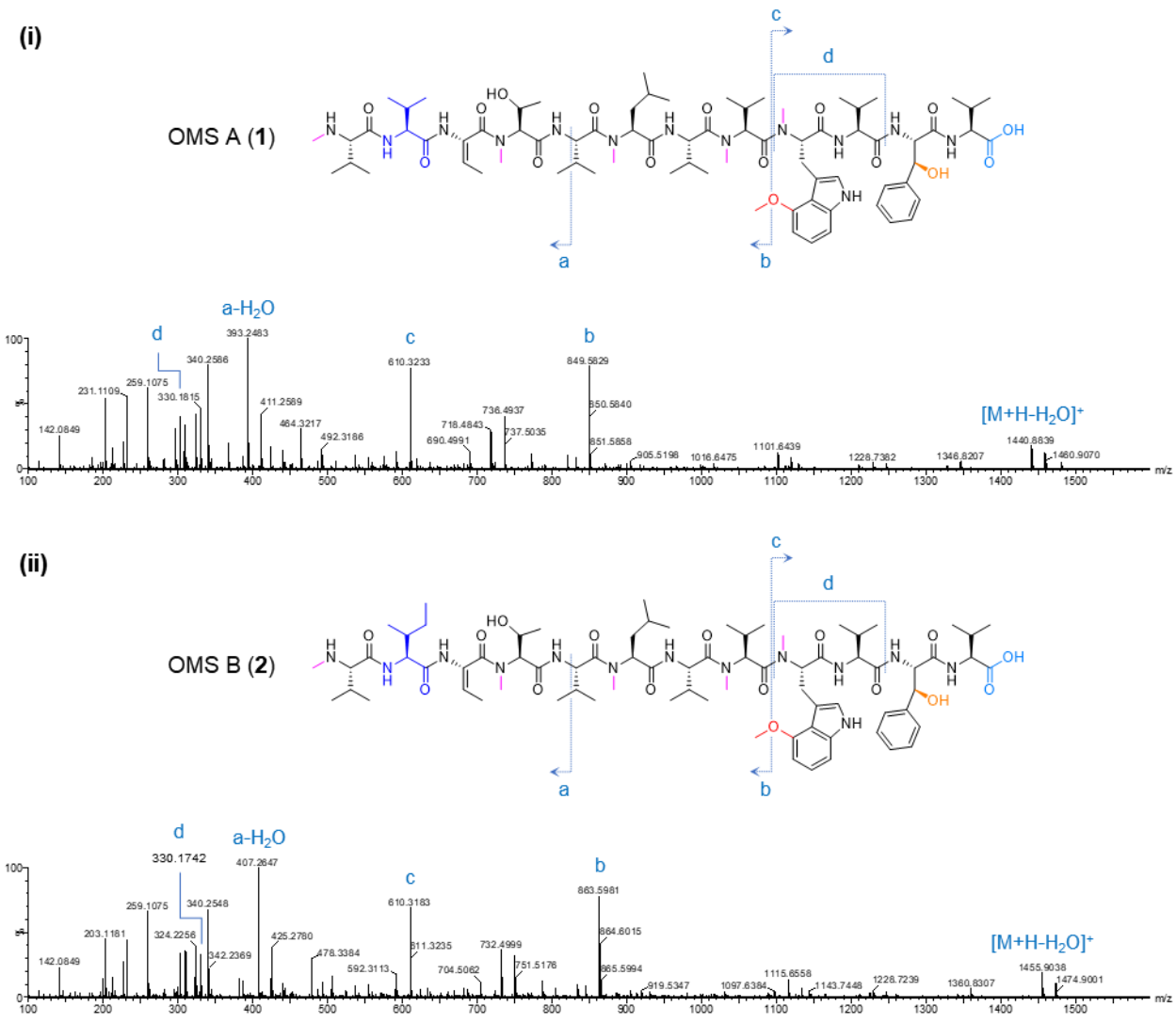


Figure S4B. Structural assignment of dehydroxylated OMS derivatives **4** and **5** produced from *ohmL* deletion mutant strain.

(i) MS/MS fragmentation pattern and MS/MS spectra of dehydroxy-OMS A (**4**) from *ΔohmL* strain.
(ii) MS/MS fragmentation pattern and MS/MS spectra of dehydroxy-OMS B (**5**) from *ΔohmL* strain.

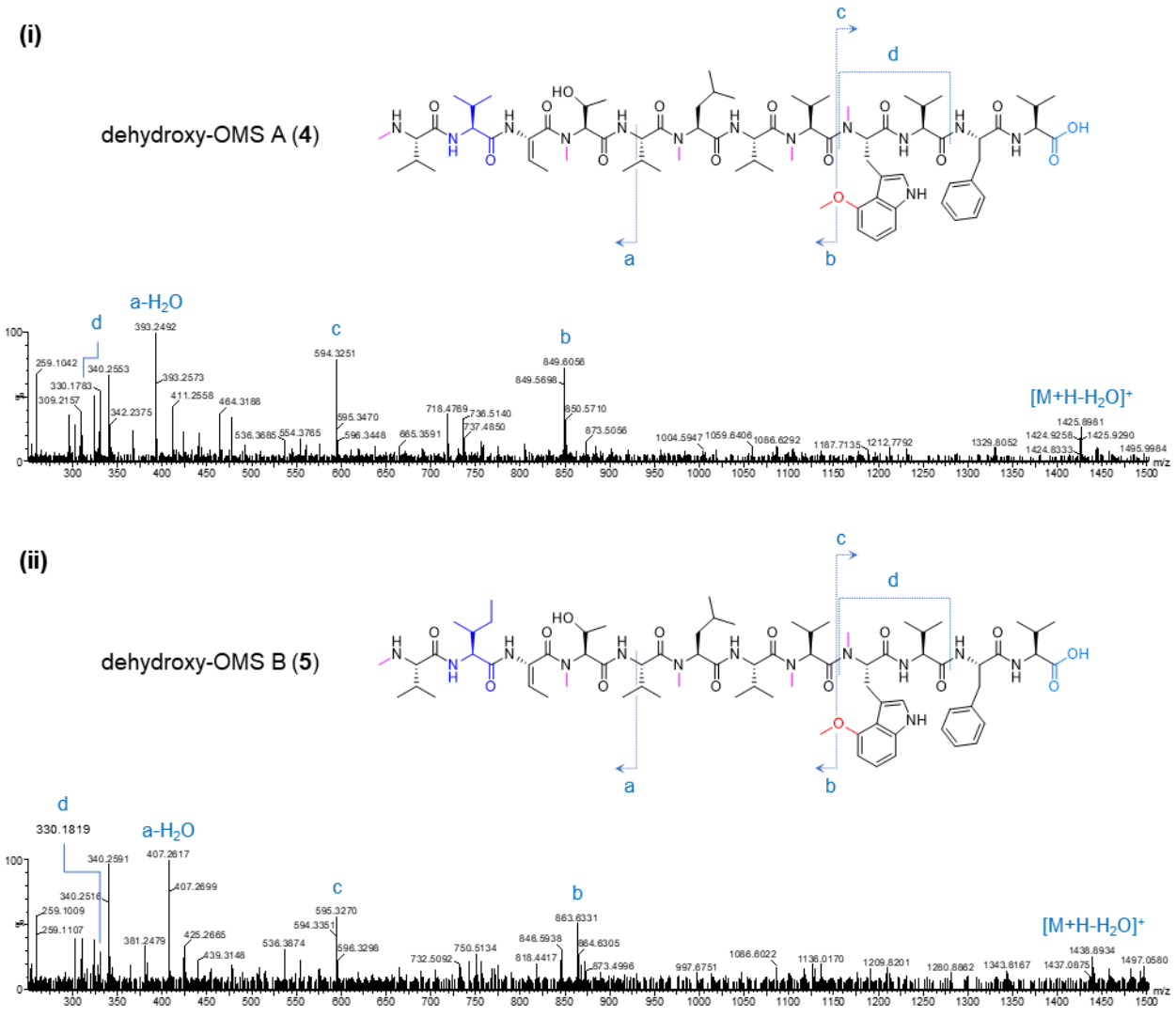


Figure S4C. Structural assignment of demethoxylated OMS derivatives **6** and **7** produced from *ohmK* deletion mutant strain.

(i) MS/MS fragmentation pattern and MS/MS spectra of demethoxy-OMS A (**6**) from *ΔohmK* strain.
(ii) MS/MS fragmentation pattern and MS/MS spectra of demethoxy-OMS B (**7**) from *ΔohmK* strain.

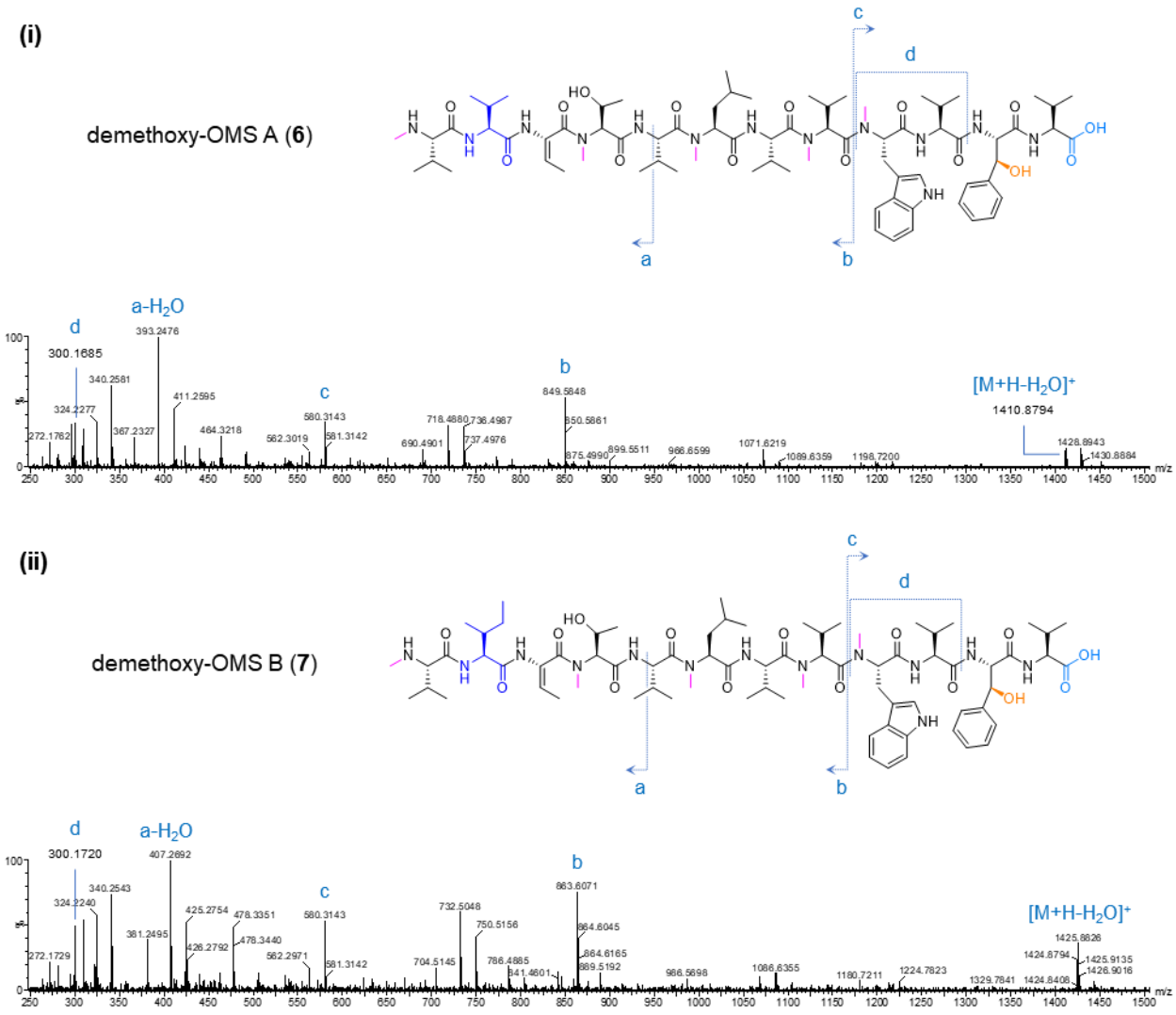


Figure S4D. Structural assignment of demethoxylated OMS derivatives **6** and **7** produced from *ohmJ* deletion mutant strain.

(i) MS/MS fragmentation pattern and MS/MS spectra of demethoxy-OMS A (**6**) from *ΔohmJ* strain.
(ii) MS/MS fragmentation pattern and MS/MS spectra of demethoxy-OMS B (**7**) from *ΔohmJ* strain.

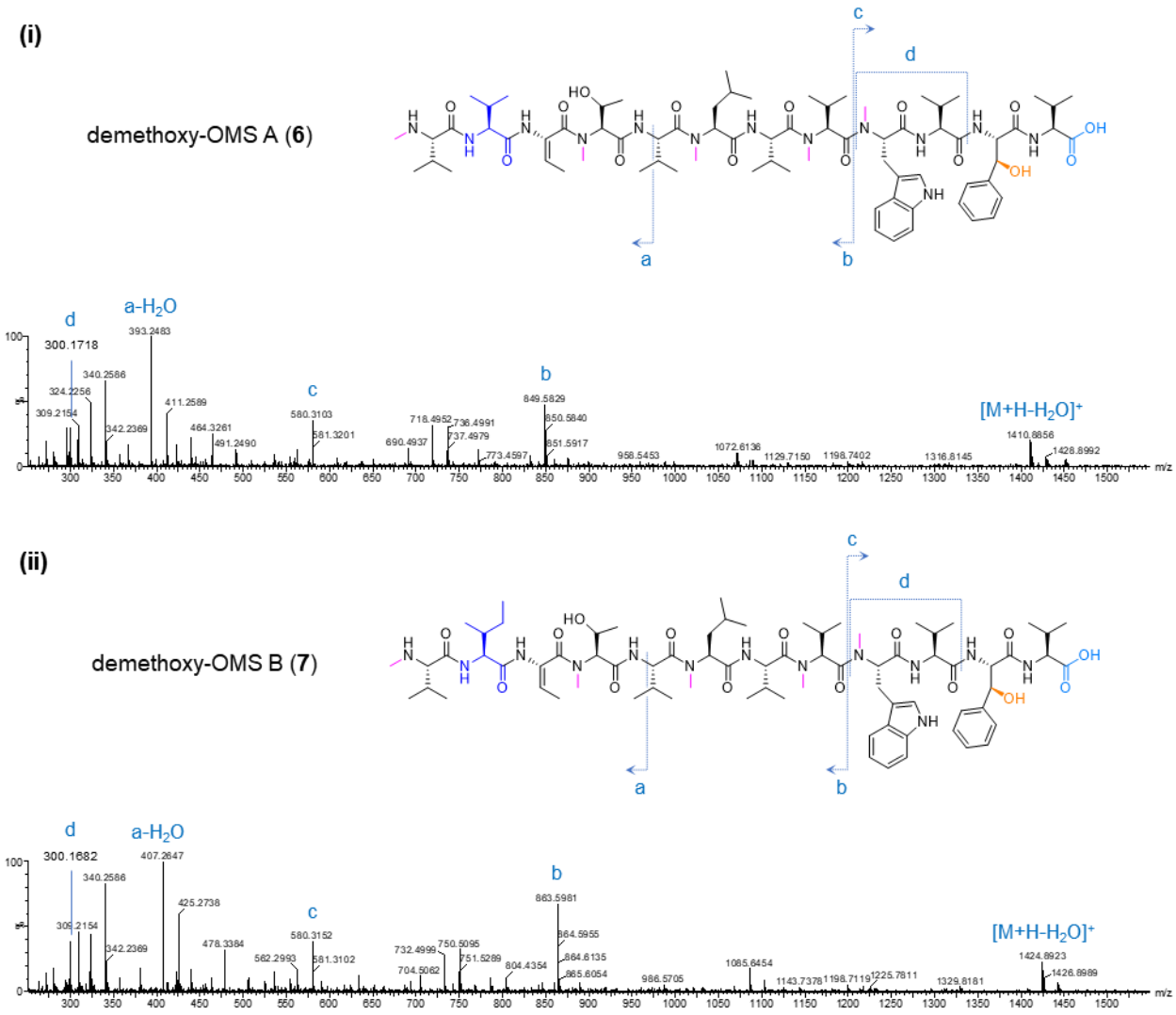


Figure S5. HR-FAB-MS data of **4**.

[Elemental Composition] Page: 1
Data : FAB-S236 Date : 15-Mar-2019 21:35
Sample: 865.1441
Note : m-NBA
Inlet : Direct Ion Mode : FAB+
RT : 1.80 min Scan#: (19,149)
Elements : C 100/0, H 200/0, N 15/10, O 20/10
Mass Tolerance : 20ppm, 5mmu if m/z < 250, 10mmu if m/z > 500
Unsaturation (U.S.) : -0.5 - 50.0

Observed m/z	Int%	Err[ppm / mmu]	U.S.	Composition
1442.9027	100.0	-4.6 / -6.7	31.0	C 83 H 118 N 12 O 10
		+4.1 / +5.9	31.5	C 82 H 116 N 13 O 10
		+3.2 / +4.6	31.0	C 84 H 118 N 10 O 11
		-1.8 / -2.7	27.0	C 78 H 118 N 14 O 12
		+6.9 / +9.9	27.5	C 77 H 116 N 15 O 12
		-2.8 / -4.0	26.5	C 80 H 120 N 11 O 13
		+5.9 / +8.6	27.0	C 79 H 118 N 12 O 13
		+0.0 / +0.0	22.5	C 75 H 120 N 13 O 15
		-0.9 / -1.3	22.0	C 77 H 122 N 10 O 16
		-5.9 / -8.5	18.0	C 71 H 122 N 14 O 17
		+2.8 / +4.0	18.5	C 70 H 120 N 15 O 17
		-6.8 / -9.9	17.5	C 73 H 124 N 11 O 18
		+1.9 / +2.7	18.0	C 72 H 122 N 12 O 18
		-4.1 / -5.8	13.5	C 68 H 124 N 13 O 20
		+4.7 / +6.7	14.0	C 67 H 122 N 14 O 20

Figure S6. ^{1}H NMR spectrum data of **4** at 800 MHz in pyridine-d_5.

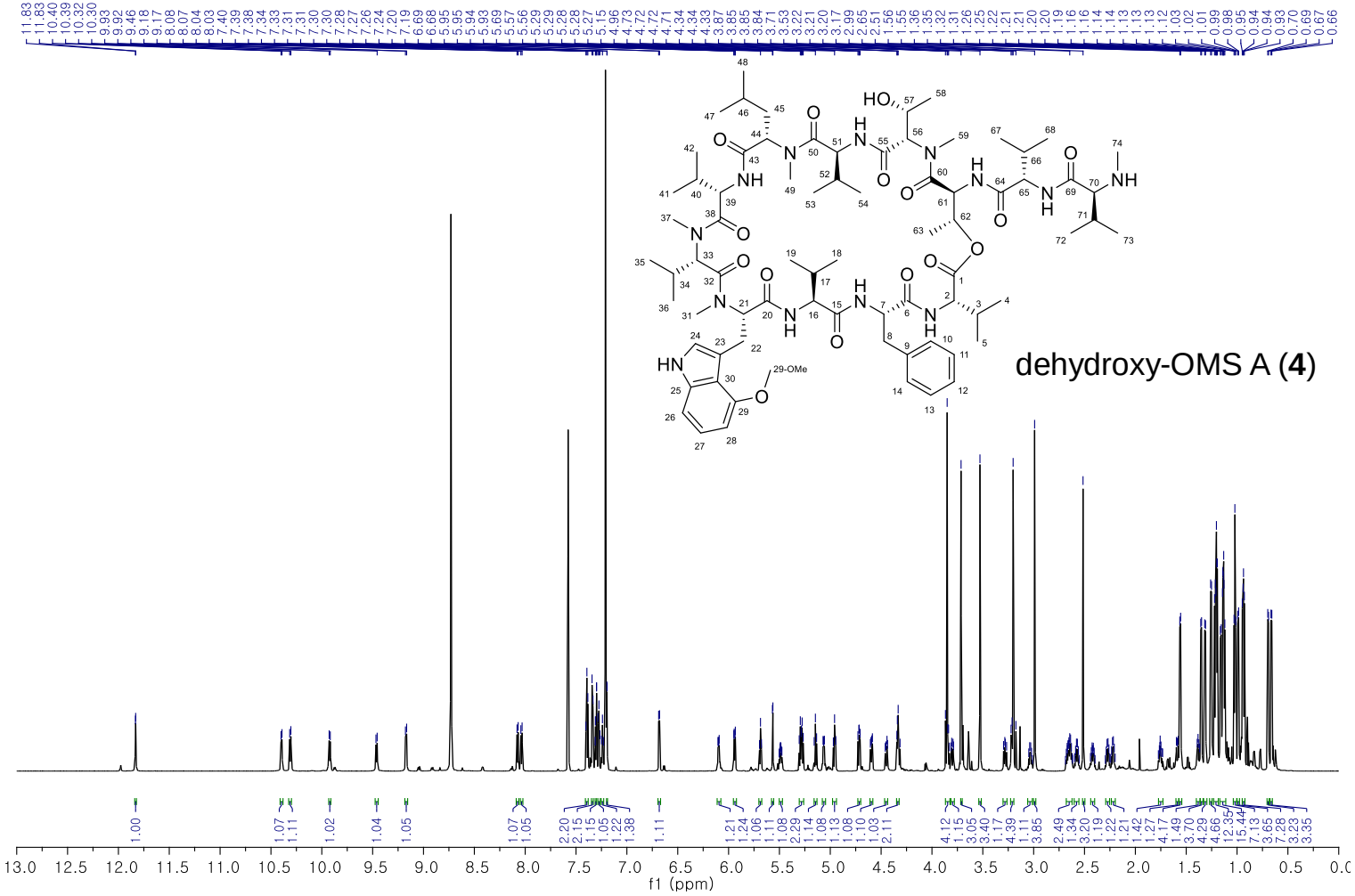


Figure S7. ^{13}C NMR spectrum data of **4** at 200 MHz in pyridine-d_5.

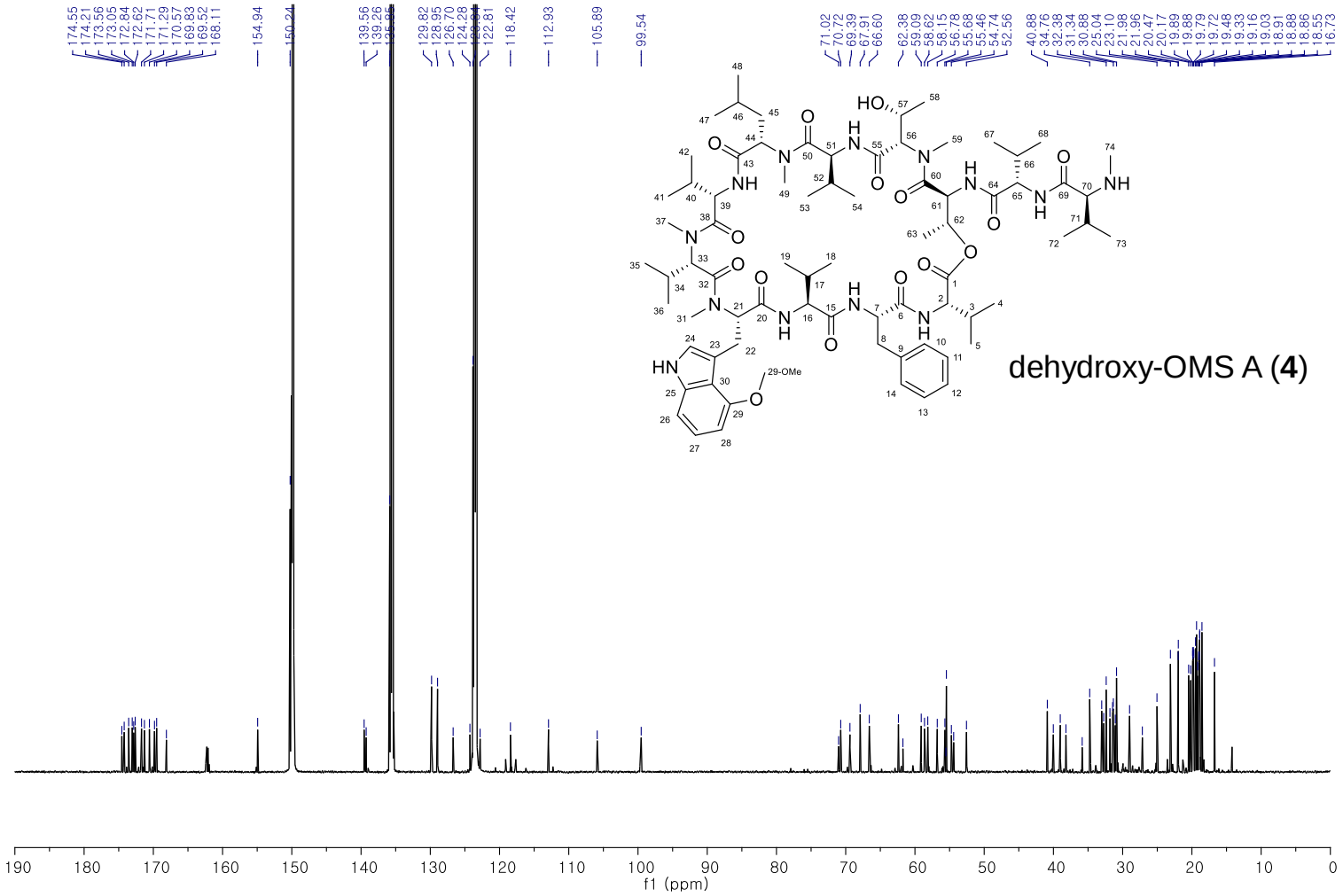


Figure S8. COSY NMR spectrum data of **4** at 800 MHz in pyridine-d_5.

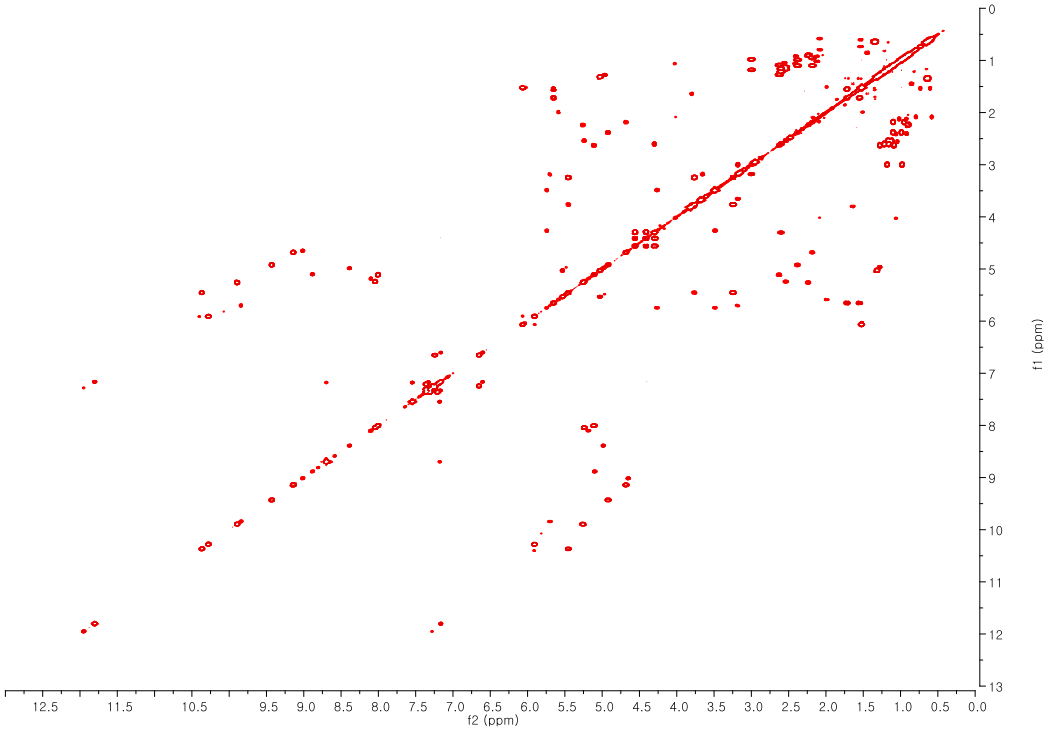


Figure S9. TOCSY NMR spectrum data of **4** at 800 MHz in pyridine-d_5.

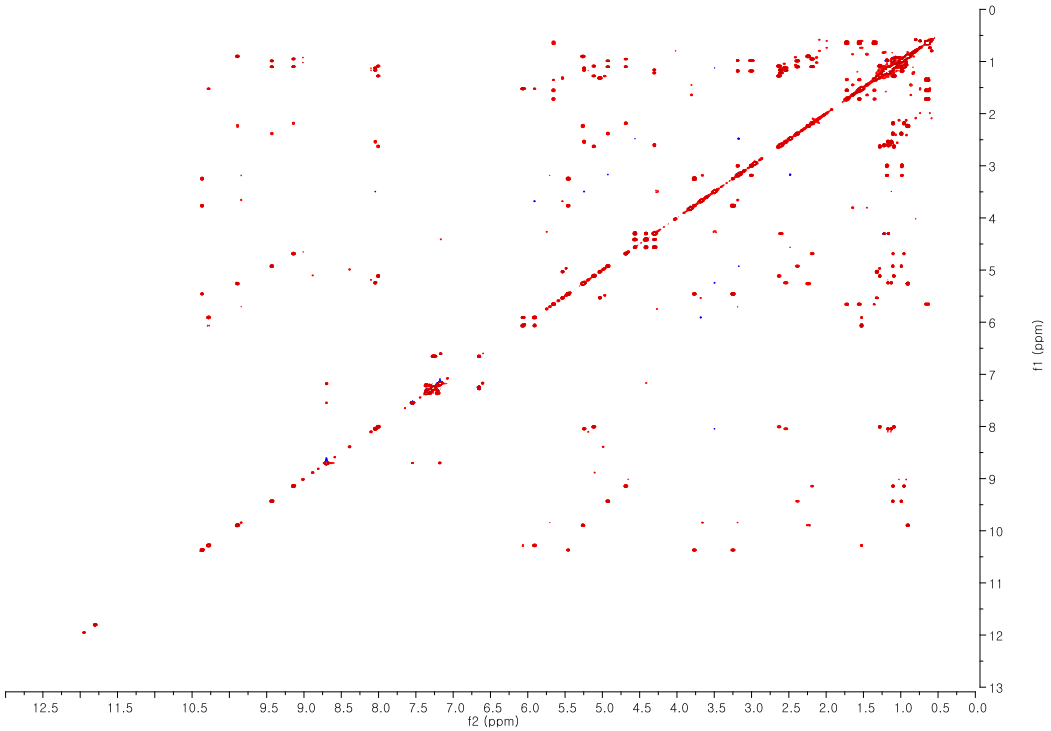


Figure S10. HSQC NMR spectrum data of **4** at 800 MHz in pyridine-d_5.

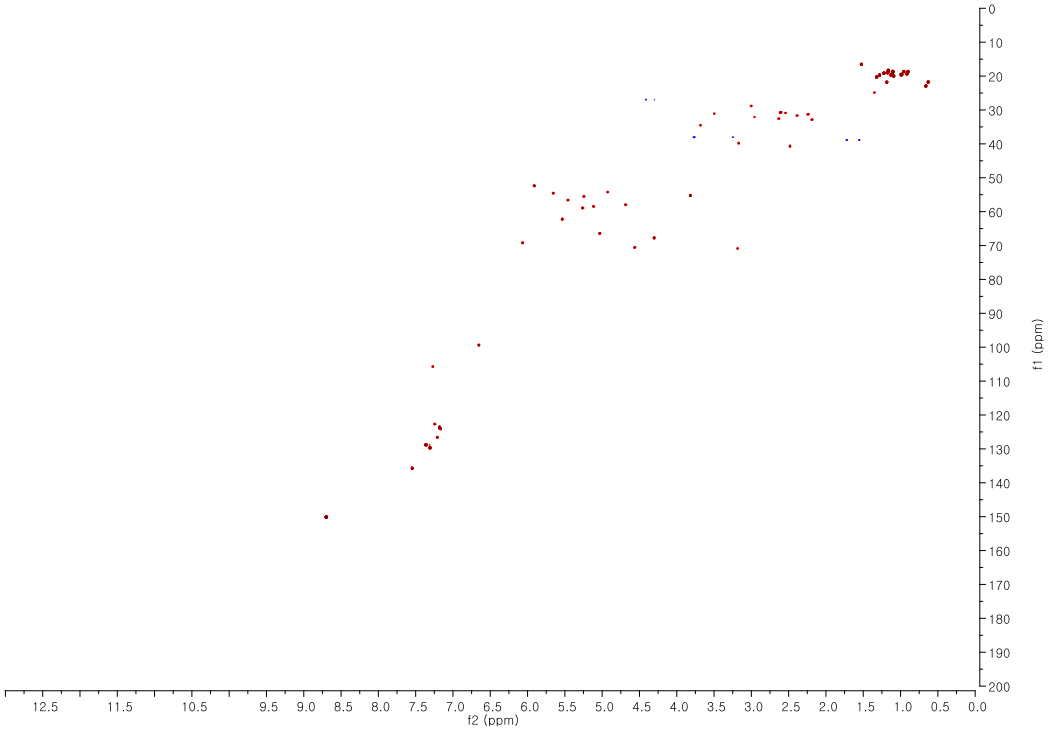


Figure S11. HMBC NMR spectrum data of **4** at 800 MHz in pyridine-d_5.

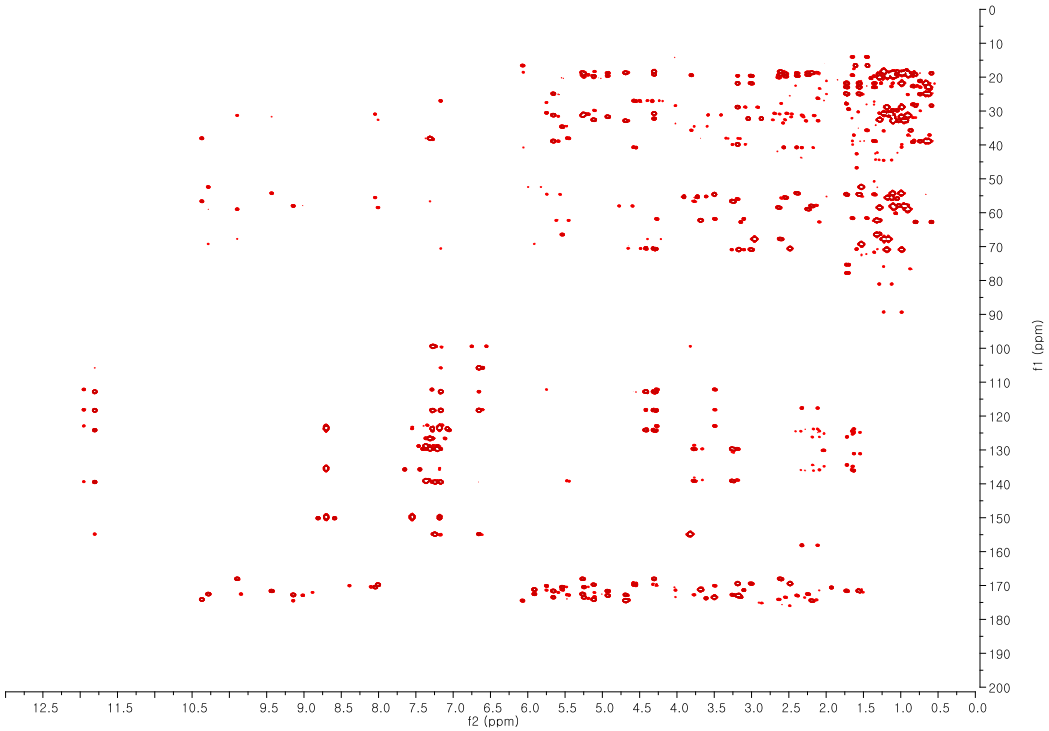


Figure S12. ROESY NMR spectrum data of **4** at 800 MHz in pyridine-d_5.

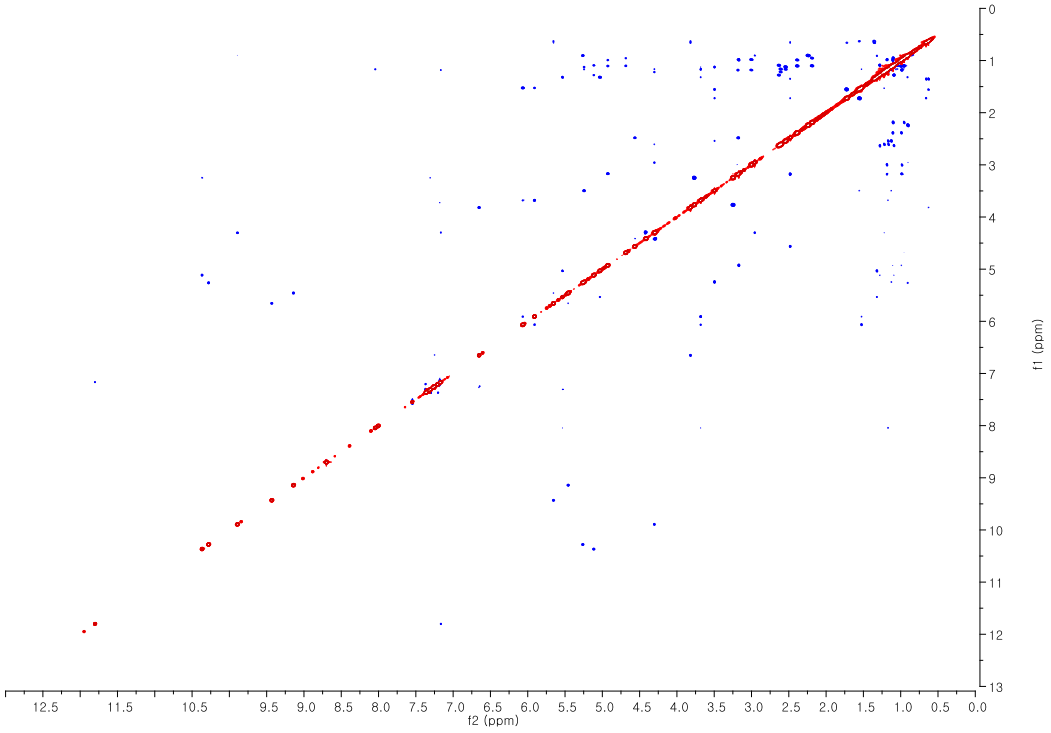


Figure S13. Comparing CD spectra data of **1**, **4**, and **6**.

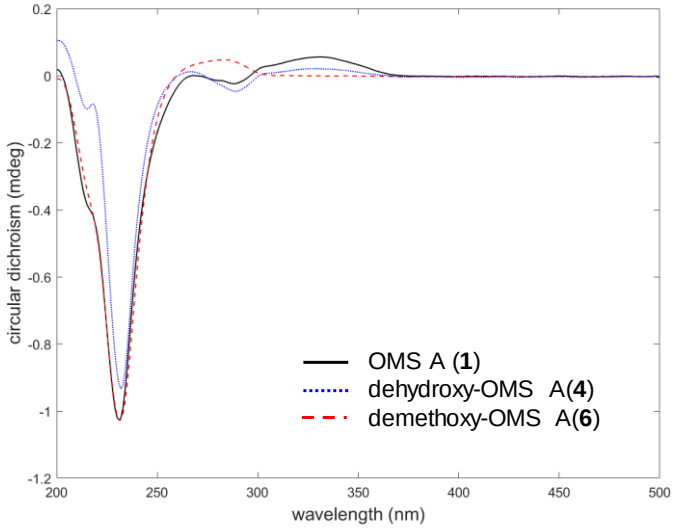
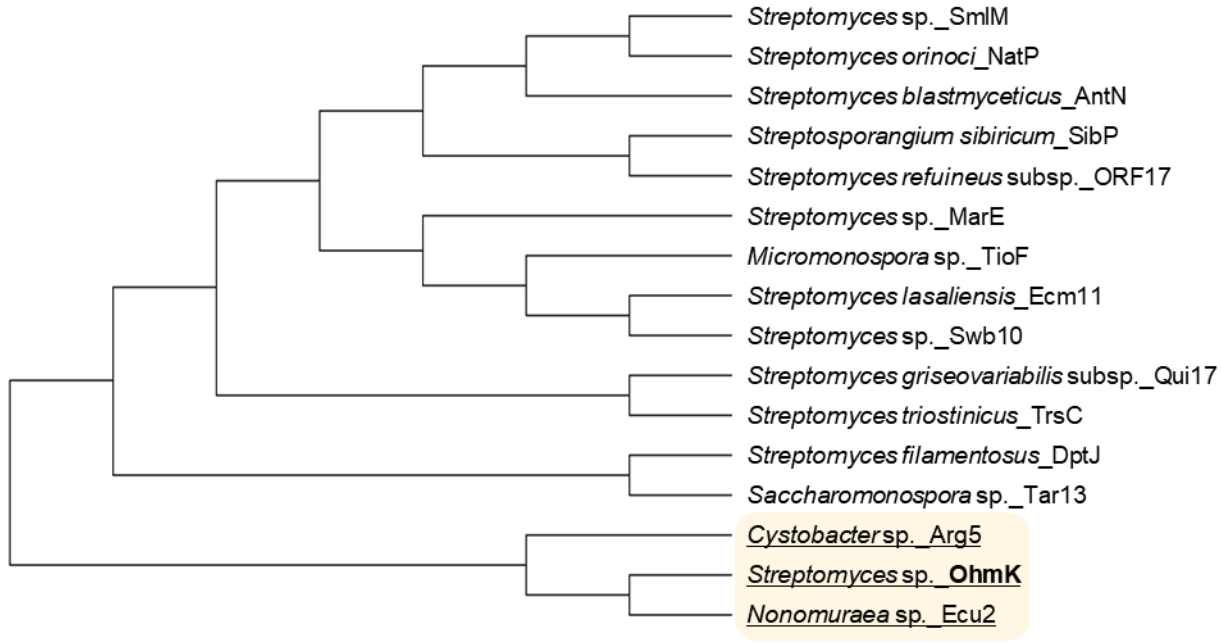


Figure S14. Phylogenetic relationships of known TDOs from secondary metabolite biosynthesis, including OhmK.



Accession numbers: SmlM (BBD17752.1, *Streptomyces* sp. ML55), NatP (BBD17773.1, *Streptomyces orinoci* NBRC 13466), AntN (AGG37776.1, *Streptomyces blastmyceticus* NBRC 12747), SibP (ACN39739.1, *Streptosporangium sibiricum* DSM 44039), ORF17 (ABW71848.1, *Streptomyces refuineus* subsp. thermotolerans), MarE (AHJ60974.1, *Streptomyces* sp. B9173), TioF (CAJ34362.1, *Micromonospora* sp. ML1), Ecm11 (BAE98160.1, *Streptomyces lasaliensis*), Swb10 (BAI63282.1, *Streptomyces* sp. SNA15896), Qui17 (AET98915.1, *Streptomyces griseovariabilis* subsp. bandungensis strain 2507), TrsC (BAH04172.1, *Streptomyces triostinicus*), DptJ (AAX31563.1, *Streptomyces filamentosus* NRRL 11379), Tar13 (AHH53511.1, *Saccharomonospora* sp. CNQ490), Arg5 (QCE43605.1, *Cystobacter* sp. SBCb004), Ecu2 (AIW58897.1, *Nonomuraea* sp. MJM5123)

Figure S15. HR-FAB-MS data of **6**.

[Elemental Composition] Page: 1
Data : FAB-S235 Date : 15-Mar-2019 21:24
Sample: 863.1427
Note : m-NBA
Inlet : Direct Ion Mode : FAB+
RT : 0.73 min Scan#: (34,35)
Elements : C 100/0, H 200/0, N 15/10, O 20/10
Mass Tolerance : 20ppm, 5mmu if m/z < 250, 10mmu if m/z > 500
Unsaturation (U.S.) : -0.5 - 50.0

Observed m/z	Int%	Err[ppm / mmu]	U.S.	Composition
1428.8860	56.8	-5.4 / -7.8	31.0	C 82 H 116 N 12 O 10
		+3.4 / +4.8	31.5	C 81 H 114 N 13 O 10
		+2.4 / +3.5	31.0	C 83 H 116 N 10 O 11
		-2.6 / -3.7	27.0	C 77 H 116 N 14 O 12
		+6.2 / +8.8	27.5	C 76 H 114 N 15 O 12
		-3.5 / -5.1	26.5	C 79 H 118 N 11 O 13
		+5.3 / +7.5	27.0	C 78 H 116 N 12 O 13
		-0.7 / -1.0	22.5	C 74 H 118 N 13 O 15
		-1.7 / -2.4	22.0	C 76 H 120 N 10 O 16
		-6.7 / -9.6	18.0	C 70 H 120 N 14 O 17
		+2.1 / +3.0	18.5	C 69 H 118 N 15 O 17
		+1.1 / +1.6	18.0	C 71 H 120 N 12 O 18
		-4.8 / -6.9	13.5	C 67 H 122 N 13 O 20
		+4.0 / +5.7	14.0	C 66 H 120 N 14 O 20

Figure S16. ^{1}H NMR spectrum data of **6** at 850 MHz in pyridine-d_5.

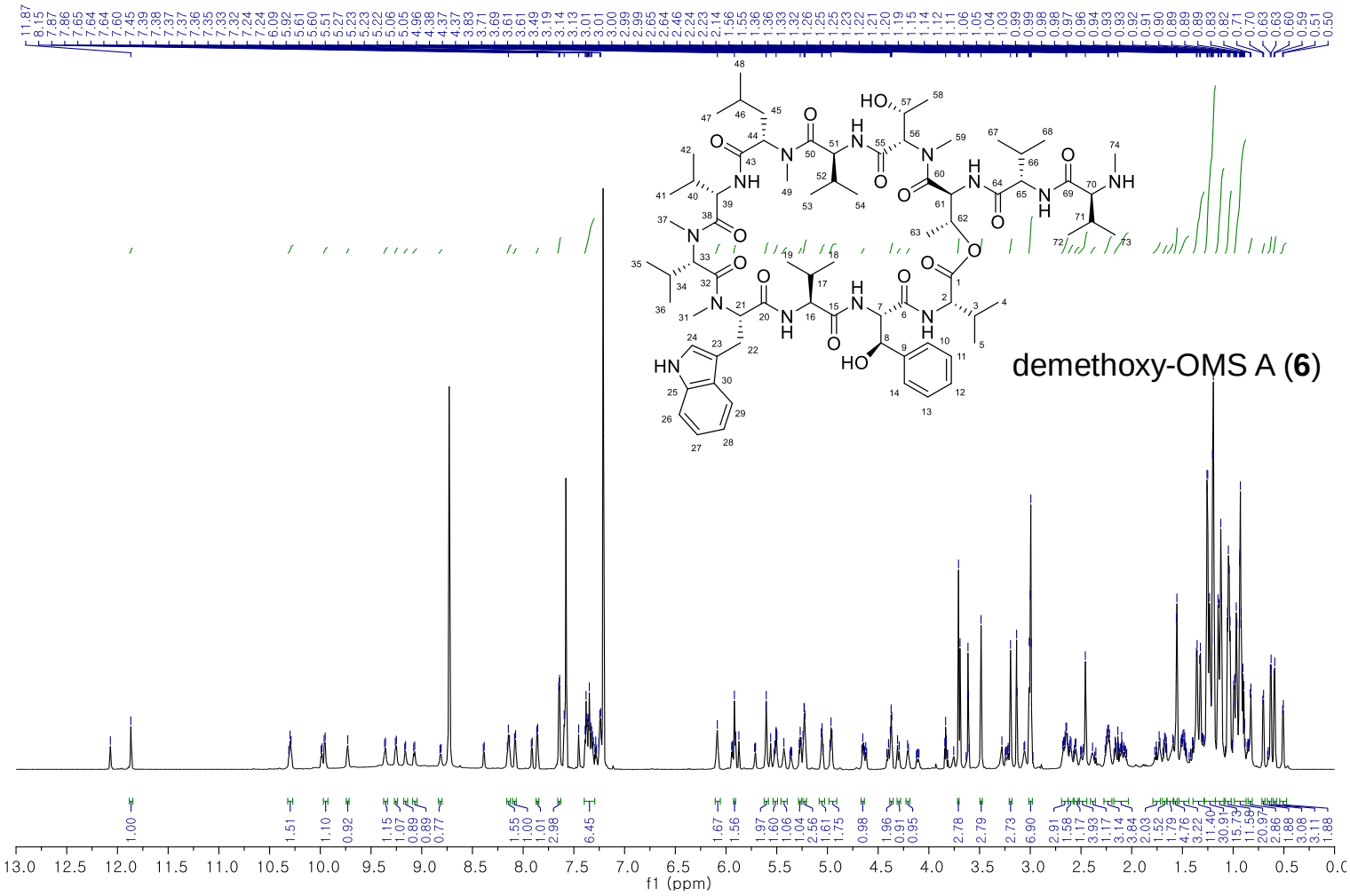


Figure S17. ^{13}C NMR spectrum data of **6** at 212.5 MHz in pyridine-d_5.

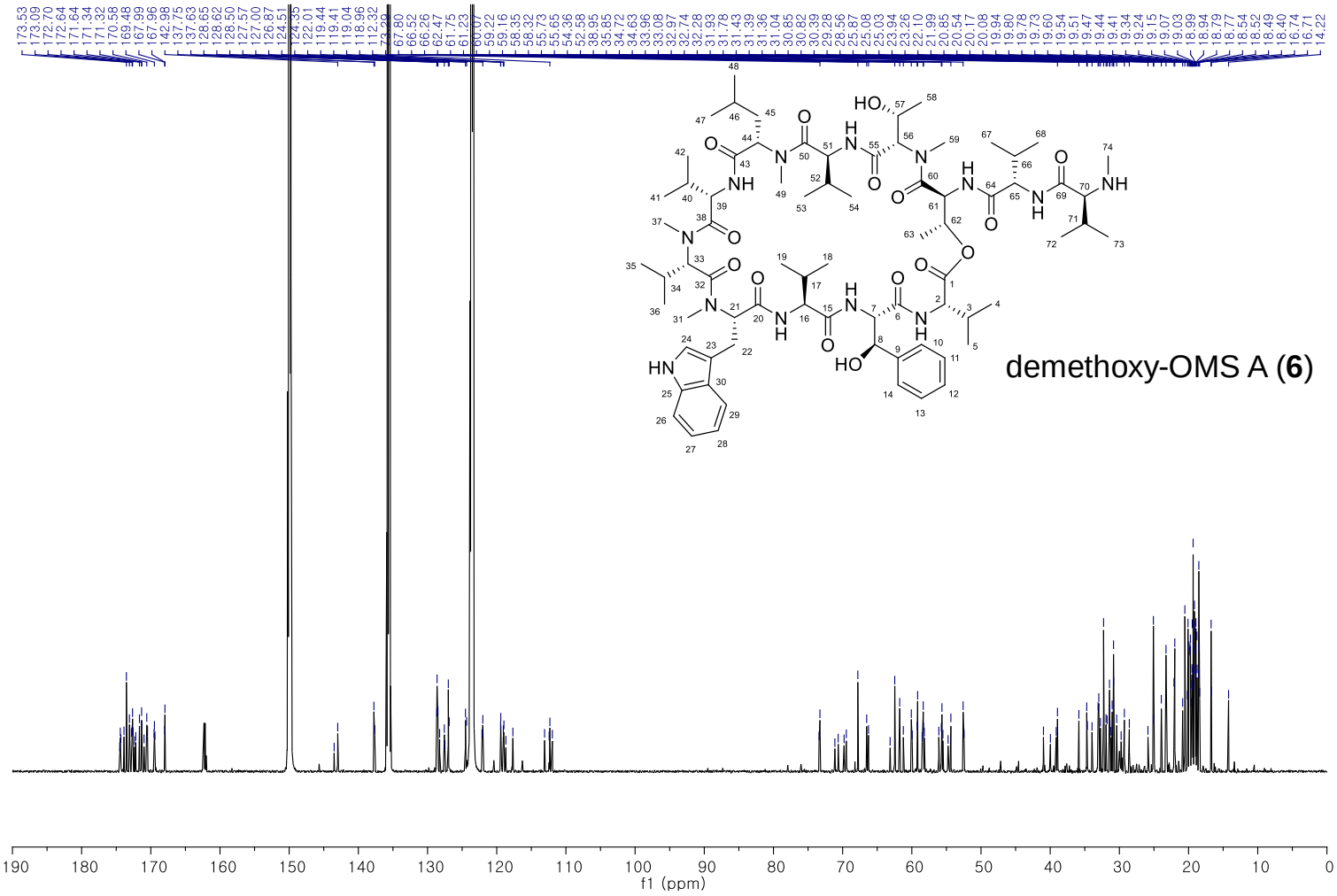


Figure S18. COSY NMR spectrum data of **6** at 850 MHz in pyridine-d_5.

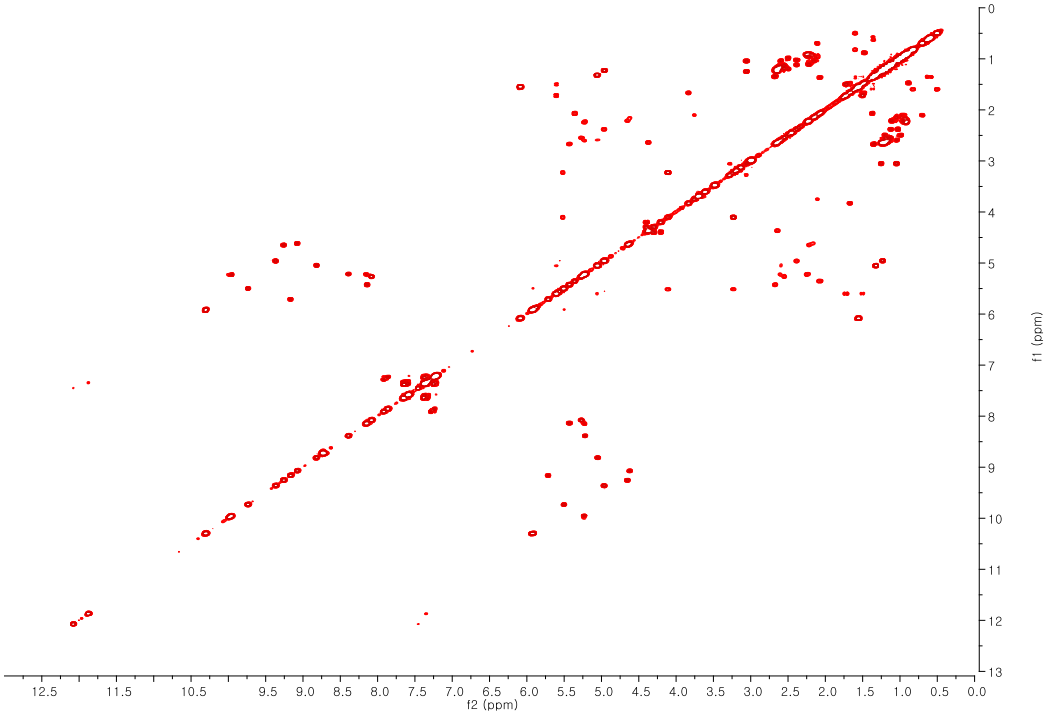


Figure S19. TOCSY NMR spectrum data of **6** at 850 MHz in pyridine-d_5.

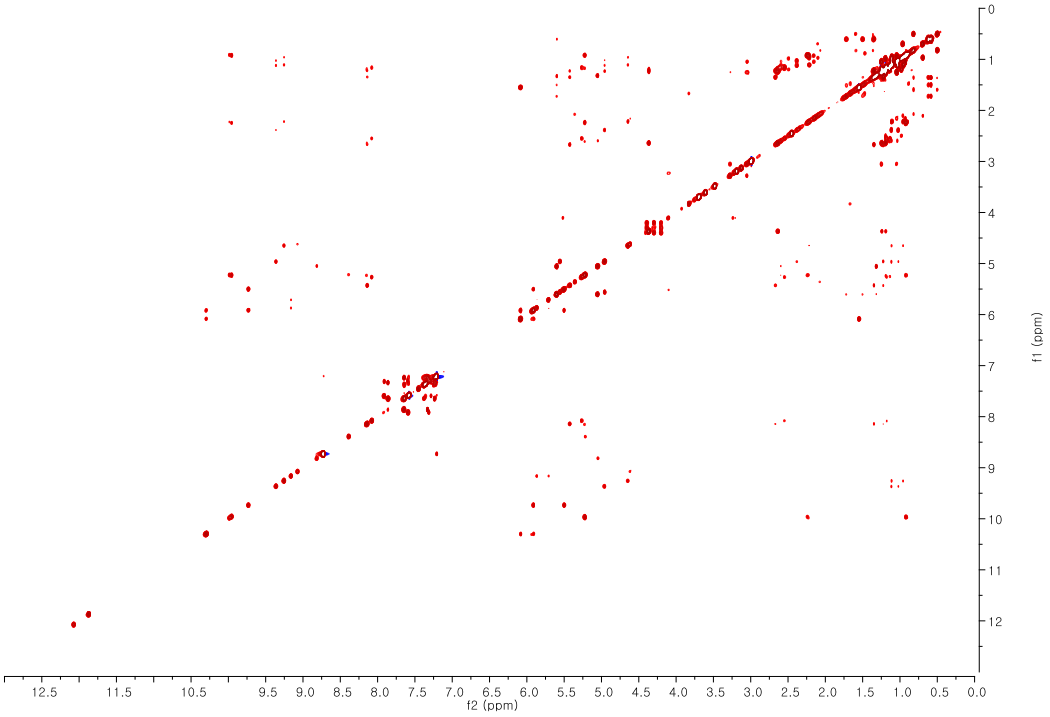


Figure S20. HSQC NMR spectrum data of **6** at 850 MHz in pyridine-d_5.

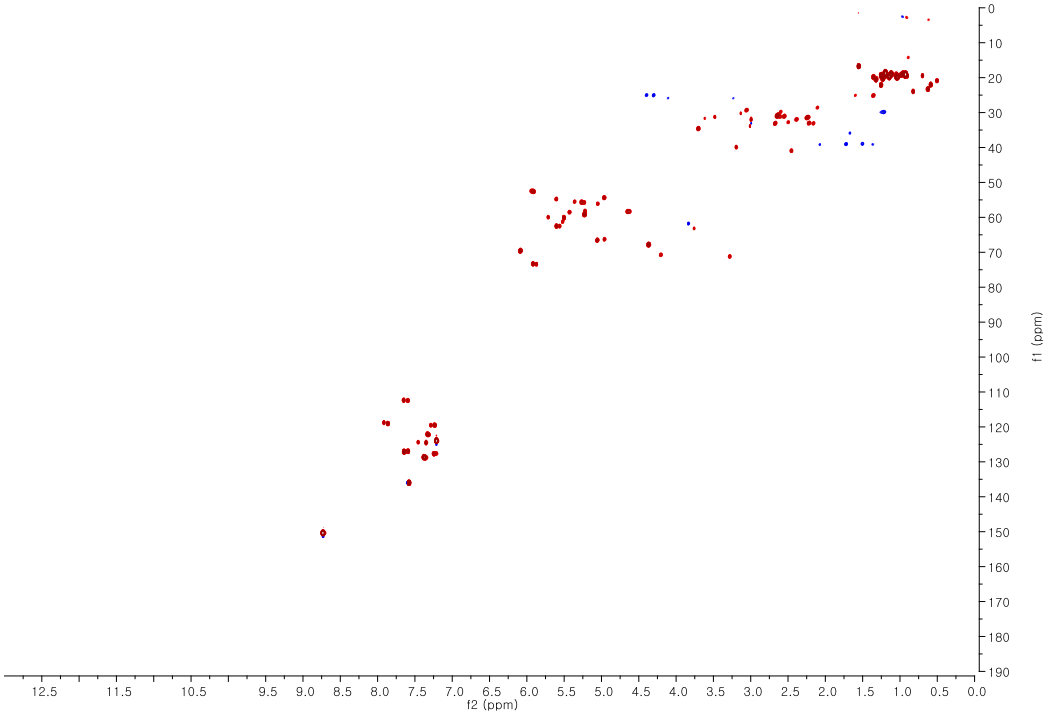


Figure S21. HMBC NMR spectrum data of **6** at 850 MHz in pyridine-d_5.

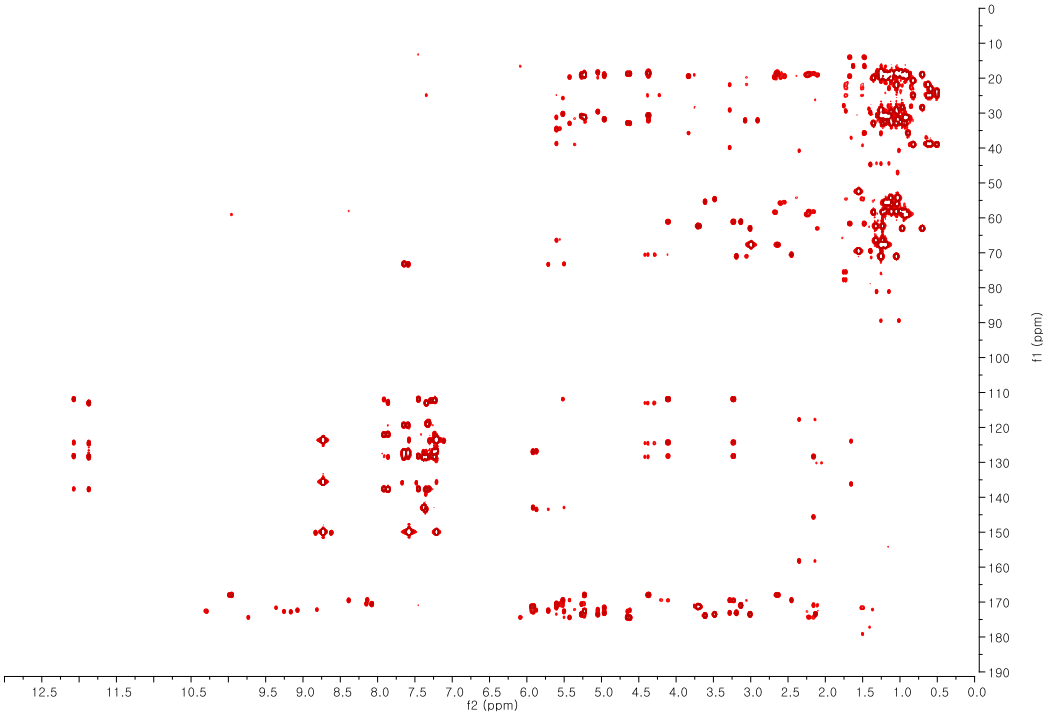


Figure S22. ROESY NMR spectrum data of **6** at 850 MHz in pyridine-d_5.

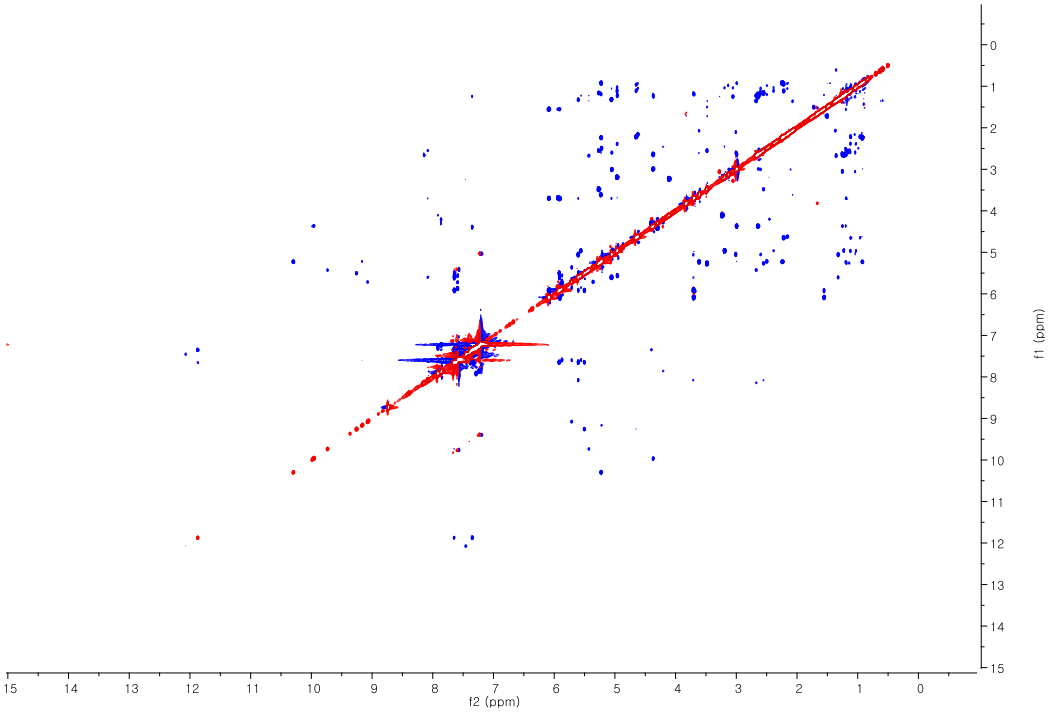


Table S1. Deduced function of ORFs in the ohmyungsamycin biosynthetic gene cluster.

ORF	Size (aa)	Proposed function	Homologue in *Nonomuraea* sp. MJM5123	
			Protein / GenBank (residues)	Identity/ Similarity
Orf(-2)	200	Hypothetical protein	-	-
Orf(-1)	291	Transposase	-	-
OhmA	13825	Nonribosomal peptide synthetase	Ecu11 / AIW58892.1 (15259)	64/74
OhmB	73	MbtH-like protein	Ecu10 / AIW58893.1 (72)	76/87
OhmC	78	Hypothetical protein	Ecu9 / AIW58894.1 (78)	77/84
OhmD	362	Putative ATPase AAA	Ecu8 / AIW58895.1 (362)	78/86
OhmE	436	Hypothetical protein	Ecu7 / AIW58902.1 (431)	78/86
OhmF	276	Hypothetical protein	Ecu6 / AIW58901.1 (233)	60/75
OhmG	400	Transposase	-	-
OhmH	320	ABC transporter ATP-binding protein	Ecu5 / AIW58900.1 (291)	72/84
OhmI	277	ABC-2 type transporter	Ecu4 / AIW58899.1 (276)	75/85
OhmJ	333	*O*-Methyltransferase	Ecu3 / AIW58898.1 (333)	61/73
OhmK	359	Putative tryptophan 2, 3-dioxygenase	Ecu2 / AIW58897.1 (368)	52/62
OhmL	398	Cytochrome P450 monooxygenase	Ecu1 / AIW58896.1 (447)	71/81
Orf(+1)	476	Hypothetical protein	-	-
Orf(+2)	327	Acetyl xylan esterase	-	-

Table S2. Substrate specificity sequences for adenylation (A) domains from ohmyungsamycin NRPS.

A domain	Substrate specific sequence	Predicted substrate	Postulated substrate
A1	D A Y W W G G T	Val	L-Val
A2	D A Y W G G A T	Val	L-Val/L-Ile
A3	D F W N V G M V	Thr	L-Thr
A4	D F W N I G M V	Thr	L-Thr
A5	D A Y W W G G T	Val	L-Val
A6	D A L L I G A I	Phe/Trp	L-Leu
A7	D A Y W W G G T	Val	L-Val
A8	D A Y W W G G T	Val	L-Val
A9	D V A L V G A V	Trp	4-MeO-L-Trp
A10	D A Y W W G G T	Val	L-Val
A11	D A W T V A A V	Phe	β-OH-L-Phe
A12	D A Y W W G G T	Val	L-Val

Table S3. Bacterial strains and plasmids used in this study.

Strain and plasmid	Description	Reference
Strain		
E. coli DH5α	Host for general cloning	New England Biolabs
E. coli ET12567/pUZ8002	Methylation-deficient donor strain for conjugal transfer between *E. coli* and *Streptomyces*	[1]
Streptomyces sp. SNJ042	Wild type ohmyungsamycin-producing strain	[2]
ΔohmL	The *ohmL* gene deleted mutant of SNJ042	This study
ΔohmK	The *ohmK* gene deleted mutant of SNJ042	This study
ΔohmJ	The *ohmJ* gene deleted mutant of SNJ042	This study
ΔohmK/ohmK	The *ohmK* gene complement mutant of *ΔohmK*	This study
ΔohmJ/ohmJ	The *ohmJ* gene complement mutant of *ΔohmJ*	This study
ΔohmJ/ohmK	The *ohmK* gene complement mutant of *ΔohmJ*	This study
Plasmid		
pKC1139	Temperature-sensitive *E. coli-Streptomyces* shuttle vector for gene disruption; *oriT* and Apr^R	[3]
pSET152	Integrative *E. coli-Streptomyces* shuttle vector containing P*ermE** for gene expression; *oriT*, *attP*, ΦC31 *int*, and Apr^R	[3]
pDel-OhmL	pKC1139 derivative for in-frame deletion of *ohmL* gene	This study
pDel-OhmK	pKC1139 derivative for in-frame deletion of *ohmK* gene	This study
pDel-OhmJ	pKC1139 derivative for in-frame deletion of *ohmJ* gene	This study
pOhmK	pSET152 derivative for expression of *ohmK* gene	This study
pOhmJ	pSET152 derivative for expression of *ohmJ* gene	This study

Table S4. Primers used in this study.

Gene	Primer	Sequences (5'-3')	Restriction site
In-frame deletion			
ohmL (P450)	delL_LF	AAAGAATTCGCGATGTGGCAGCTCAGTTT	*Eco*RI
	delL_LR	AATCTAGATGAGCGGGCGGAACGGCGCG	*Xba*I
	delL_RF	AAATCTAGAGACCACGTGATCGCTCCCCT	*Xba*I
	delL_RR	TTAAGCTTGATCTATCTGCTCGCCTTCC	*Hind*III
ohmK (TDO)	delK_LF	AATTGAATTCATGGTGAACTCGCTCCTCGA	*Eco*RI
	delK_LR	AATCTAGACACATTCCCCCTGGAACGTG	*Xba*I
	delK_RF	TTAATCTAGAGTGCACTTCGCCTACCGCTG	*Xba*I
	delK_RR	TTAAGCTTGCTGAGCGGGAGATTCCTCC	*Hind*III
ohmJ (O-MT)	delJ_LF	AATCAATTGAGTGCAGAACCTCCAGATGT	*Mfe*I
	delJ_LR	AAATCTAGACATTCTCCACCCCCAGTGAA	*Xba*I
	delJ_RF	AATCTAGATGACACGTTCCAGGGGGAAT	*Xba*I
	delJ_RR	TTAAGCTTACGAGGAGATCTTCCTCAAC	*Hind*III
Complementation			
ohmK (TDO)	ohmK_F	AATTAGATCTTGACACGTTCCAGGGGGAAT	*Bgl*II
	ohmK_R	AATCTAGAGCCTCAGCGGTAGGCGAAGT	*Xba*I
ohmJ (O-MT)	ohmJ_F	AATTAGATCTTCGCTCGGGTAGTGTCACCT	*Bgl*II
	ohmJ_R	AATCTAGAGTGTCAGGGCTTGTGGGCCA	*Xba*I

Table S5. ^{1}H NMR data for minor conformer of **4** in pyridine-d_5.

	position	type	δ_H		position	type	δ_H
Val-1	1	C		Val-6	38	C	
	2	CH	4.69		39	CH	5.13
	2-NH		9.05		39-NH		8.91
	3	CH	2.15		40	CH	2.59
	4	CH_3	1.06		41	CH_3	1.16
	5	CH_3	0.95		42	CH_3	1.08
Phe-2	6	C		*N*-Me-	43	C	
	7	CH	5.73	Leu-7	44	CH	5.62
	7-NH		9.87		45a	CH_2	2.02
	8a	CH_2	3.68		45b		1.54
	8b		3.20		46	CH	1.56
	9	C			47	CH_3	0.77
	10	CH	7.30		48	CH_3	0.64
	11	CH	7.37		49	CH_3	3.64
	12	CH	7.21	Val-8	50	C	
	13	CH	7.37		51	CH	5.22
	14	CH	7.30		51-NH		8.13
Val-3	15	C			52	CH	2.62
	16	CH	5.02		53	CH_3	1.25
	16-NH		8.41		54	CH_3	1.14
	17	CH	2.44	*N*-Me-	55	C	
	18	CH_3	1.10	Thr-9	56	CH	5.52
	19	CH_3	0.96		57	CH	5.00
N-Me-4-	20	C			58	CH_3	1.30
methoxy-	21	CH	5.77		59	CH_3	3.70
Trp-4	22a	CH_2	4.30	Thr-10	60	C	
	22b		3.51		61	CH	5.93
	23	C			61-NH		10.30
	24	CH	7.31		62	CH	6.11
	24-NH		11.97		63	CH_3	1.55
	25	C		Val-11	64	C	
	26	CH	7.36		65	CH	5.28
	27	CH	7.20		65-NH		9.93
	28	CH	6.64		66	CH	2.27
	29	C			67	CH_3	0.94
	29-OMe	CH_3	3.87		68	CH_3	0.93
	30	C		*N*-Me-	69	C	
	31	CH_3	3.13	Val-12	70	CH	4.33
N-Me-	32	C			71	CH	2.64
Val-5	33	CH	4.04		72	CH_3	1.24
	34	CH	2.11		73	CH_3	1.18
	35	CH_3	0.83		74	CH_3	2.98
	36	CH_3	0.61				
	37	CH_3	3.17				

^{1}H NMR data were measured at 800 MHz.

Figure S23. Key ROESY correlations of (A) major and (B) minor conformers of dehydroxy-OMS A (**4**).

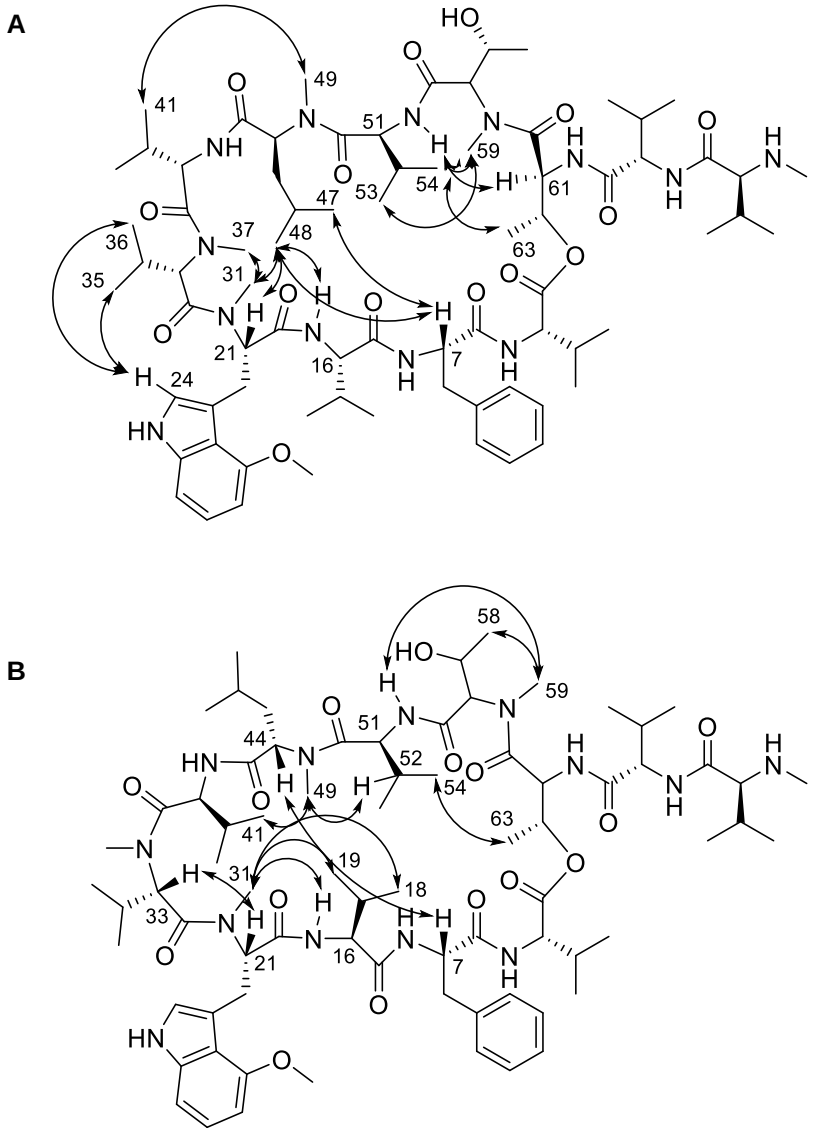
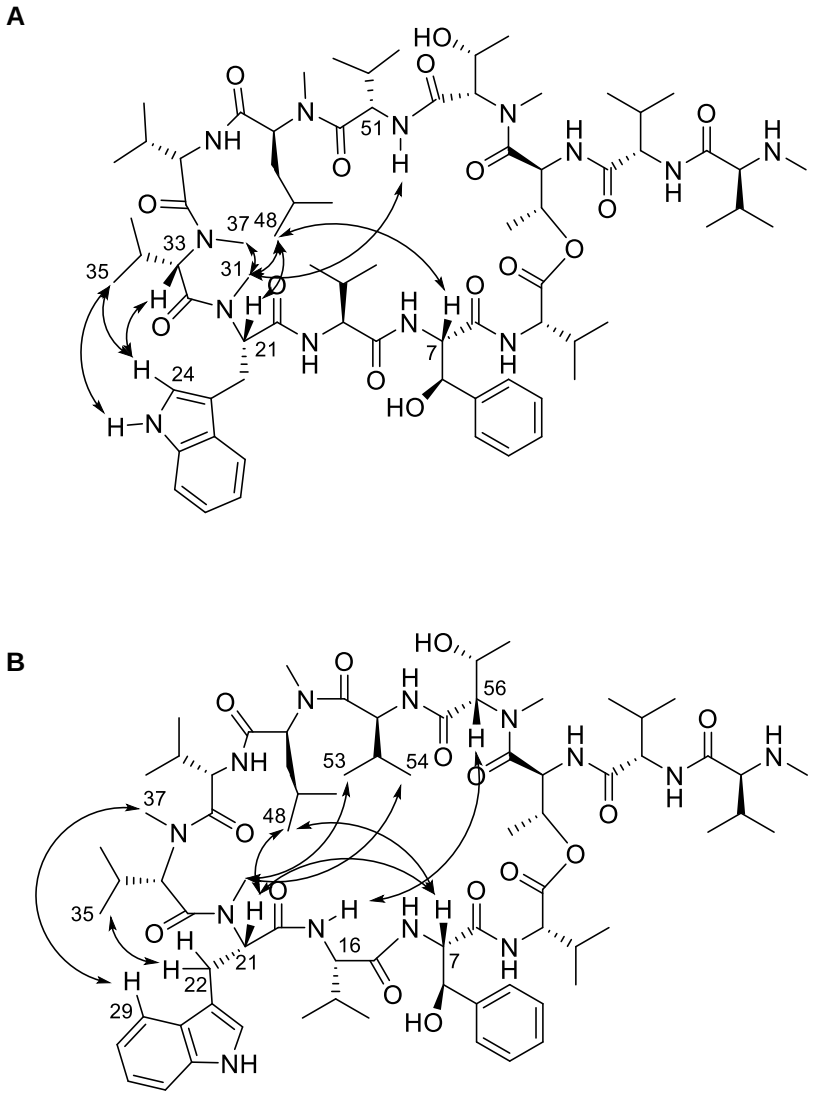


Table S6. ^{1}H NMR data for minor conformer of **6** in pyridine-d_5.

	position	type	δ_H		position	type	δ_H
Val-1	1	C		Val-6	38	C	
	2	CH	4.62		39	CH	5.05
	2-NH		9.08		39-NH		8.81
	3	CH	2.17		40	CH	2.59
	4	CH_3	0.99		41	CH_3	1.14
	5	CH_3	1.05		42	CH_3	1.04
β-OH-	6	C		*N*-Me-	43	C	
Phe-2	7	CH	5.71	Leu-7	44	CH	5.36
	7-NH		9.16		45a	CH_2	2.07
	8	CH	5.88		45b		1.39
	9	C			46	CH	1.60
	10	CH	7.59		47	CH_3	0.83
	11	CH	7.37		48	CH_3	0.50
	12	CH	7.25		49	CH_3	3.61
	13	CH	7.37	Val-8	50	C	
	14	CH	7.59		51	CH	5.22
Val-3	15	C			51-NH		8.39
	16	CH	5.23		52	CH	2.50
	16-NH		8.16		53	CH_3	1.20
	17	CH	2.23		54	CH_3	0.99
	18	CH_3	0.93	*N*-Me-	55	C	
	19	CH_3	0.93	Thr-9	56	CH	5.56
N-Me-	20	C			57	CH	4.95
Trp-4	21	CH	5.52		58	CH_3	1.25
	22a	CH_2	4.11		59	CH_3	3.69
	22b		3.23	Thr-10	60	C	
	23	C			61	CH	5.94
	24	CH	7.45		61-NH		10.30
	24-NH		12.07		62	CH	6.09
	25	C			63	CH_3	1.56
	26	CH	7.59	Val-11	64	C	
	27	CH	7.37		65	CH	5.24
	28	CH	7.29		65-NH		9.98
	29	CH	7.91		66	CH	2.24
	30	C			67	CH_3	0.92
	31	CH_3	3.14		68	CH_3	0.92
N-Me-	32	C		N-Me-	69	C	
Val-5	33	CH	3.76	Val-12	70	CH	4.37
	34	CH	2.10		71	CH	2.65
	35	CH_3	0.98		72	CH_3	1.25
	36	CH_3	0.71		73	CH_3	1.20
	37	CH_3	3.01		74	CH_3	3.00

^{1}H NMR data were measured at 850 MHz.

Figure S24. Key ROESY correlations of (A) major and (B) minor conformers of demethoxy-OMS A (**6**).



References

1. Kieser, T.; Bibb, M. J.; Buttner, M. J.; Chater, K. F.; Hoopwood, D. A. *Practical Streptomyces Genetics*; The John Innes Foundation: Norwich, UK, 2000.
2. Um, S.; Choi, T. J.; Kim, H.; Kim, B. Y.; Kim, S.-H.; Lee, S. K.; Oh, K.-B.; Shin, J.; Oh, D.-C. Ohmyungsamycins A and B: cytotoxic and antimicrobial cyclic peptides produced by *Streptomyces* sp. from a volcanic island. *J. Org. Chem.* **2013**, *78*, 12321-12329.
3. Bierman, M.; Logan, R.; O'Brien, K.; Seno, E. T.; Rao, R. N.; Schoner, B. E. Plasmid cloning vectors for the conjugal transfer of DNA from *Escherichia coli* to *Streptomyces* spp. *Gene* **1992**, 116, 43-49.