SUPPORTING INFORMATION

Plantazolicin A and B: Structure elucidation of ribosomally synthesized thiazole/oxazole peptides from *Bacillus amyloliquefaciens* FZB42

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S1. Experimental section

Fermentation and isolation

The fermentations of the Mutant RSpMarA2 from *Bacillus amyloliquefaciens* FZB42 were carried out for 24 hours on 200 plates prepared with the glucose minimal medium (ammonium sulfate, 0.2%; dipotassium phosphate, 1.4%; monopotassium phosphate, 6%; sodium citrate ($2H_2O$), 0.1%; magnesium sulfate ($7H_2O$), 0.02%; agar and glucose, 0.5%).[1] Feeding fermentations were performed by adding ^{15}N-ammonium sulfate to the media instead of ^{14}N-ammonium sulfate. The bacteria were removed from the plates and extracted with acetonitrile/water (7:3; v:v; 0.1% formic acid).[2] The sample was concentrated *in vacuo* to yield a brown solid. The crude product was dissolved in acetonitrile/water (1:1; v:v) and solid phase extraction was performed. Fractions containing **1a** or **1b** were concentrated to dryness. 2 mg of plantazolicin A were obtained as a white solid.

Structure elucidation

ESI-MS, ESI-MS-MS and high-resolution ESI-MS mass spectra were recorded using a LTQ-Orbitrap XL (Thermo Scientific, Bremen, Germany) coupled to an Agilent 1260 HPLC system (Agilent Technologies, Waldbronn, Germany). NMR experiments were performed on a DRX 500 NMR spectrometer (Bruker, Karlsruhe, Germany) equipped with a broadband inverse detection probe head with z gradients. DMSO-*d6* was used as solvents for NMR experiments and chemical shifts were referenced to the solvent peaks (δ_H 2.49 ppm, δ_C 39.7 ppm).

All ^{15}N-NMR experiments (^{15}N-HMQC/HMBC) were performed at the Leibniz Institute of Molecular Pharmacology (FMP, Berlin, Germany). ^{15}N-HMQC- and ^{15}N-HMBC-NMR experiments of plantazolicin A (**1b**) were acquired in DMSO-*d6* at 300 K on a AV 600 NMR spectrometer (Bruker, Karlsruhe, Germany) equipped with a TXI cryo probe head with z gradient or a AV 900 NMR spectrometer equipped with a TCI cryo probe head with z gradient. All chemical shifts are referenced to liquid NH_3.

Table S1. Physico-chemical properties of plantazolicin A

		plantazolicin A
Appearance		white solid
Molecular mass (g/mol)		1335.47
Molecular formula		$C_{33}H_{19}O_{13}N_{17}S_2$
High-resolution orbitrap-ESI-MS (*m/z*)	Found	1336.47612 $[M+H]^+$
	Calcd.	1336.47804, Δm -1.027 p.p.m.
UV λ_{max}^{ACN/H_2O} [nm]		267
$[\alpha]_D^{23}$ (c 0.1, ACN/H_2O)		-1.2

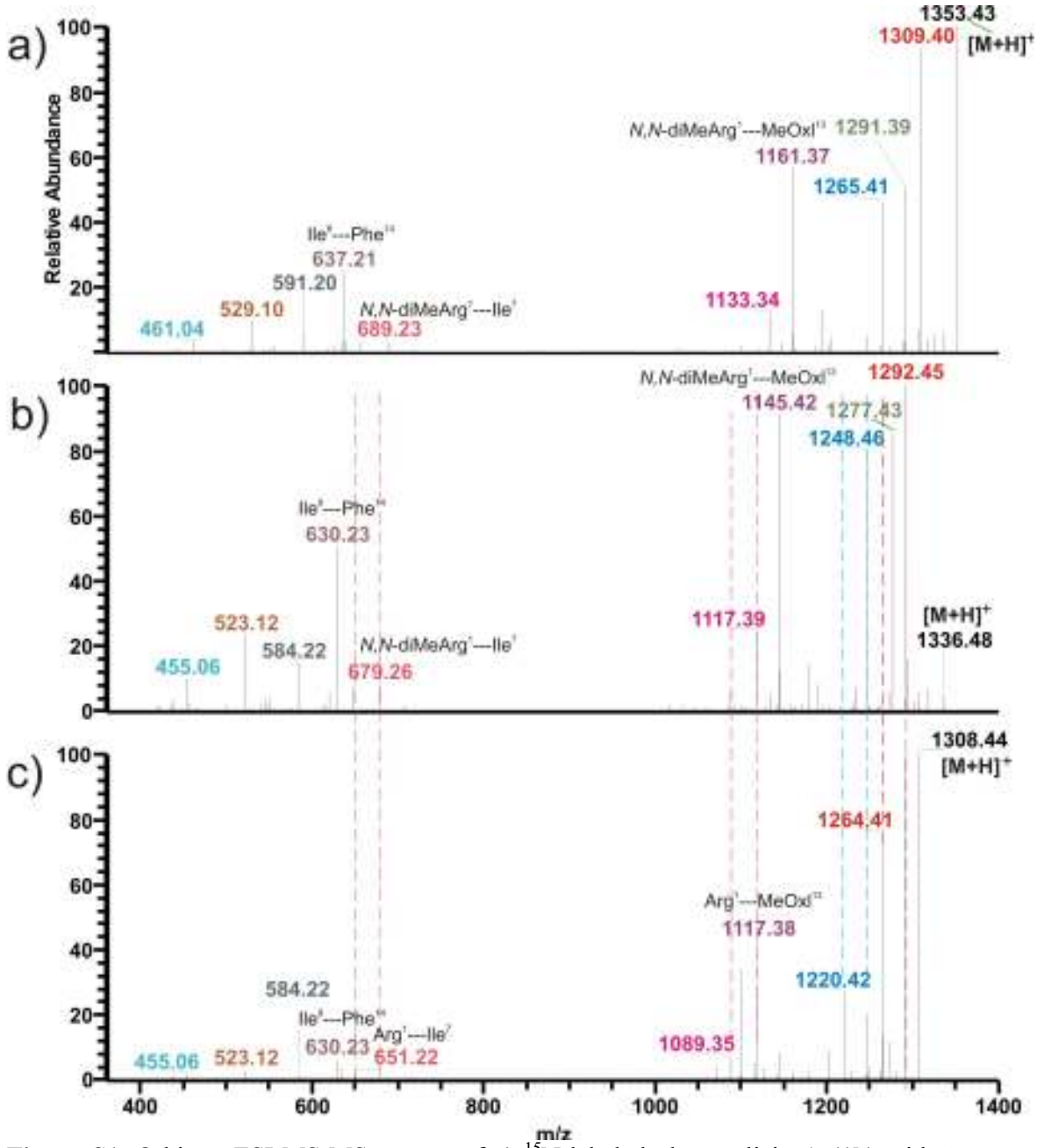

Figure S1. Orbitrap-ESI-MS-MS spectra of a) ^{15}N-labeled plantazolicin A (**1b**) with Δm = +17 amu compared to b) plantazolicin A (**1a**) and c) the desmethyl plantazolicin B (**2**). Dashed lines symbolize Δm = 28 amu between **1a** and **2** corresponding to the lack of two methyl groups.

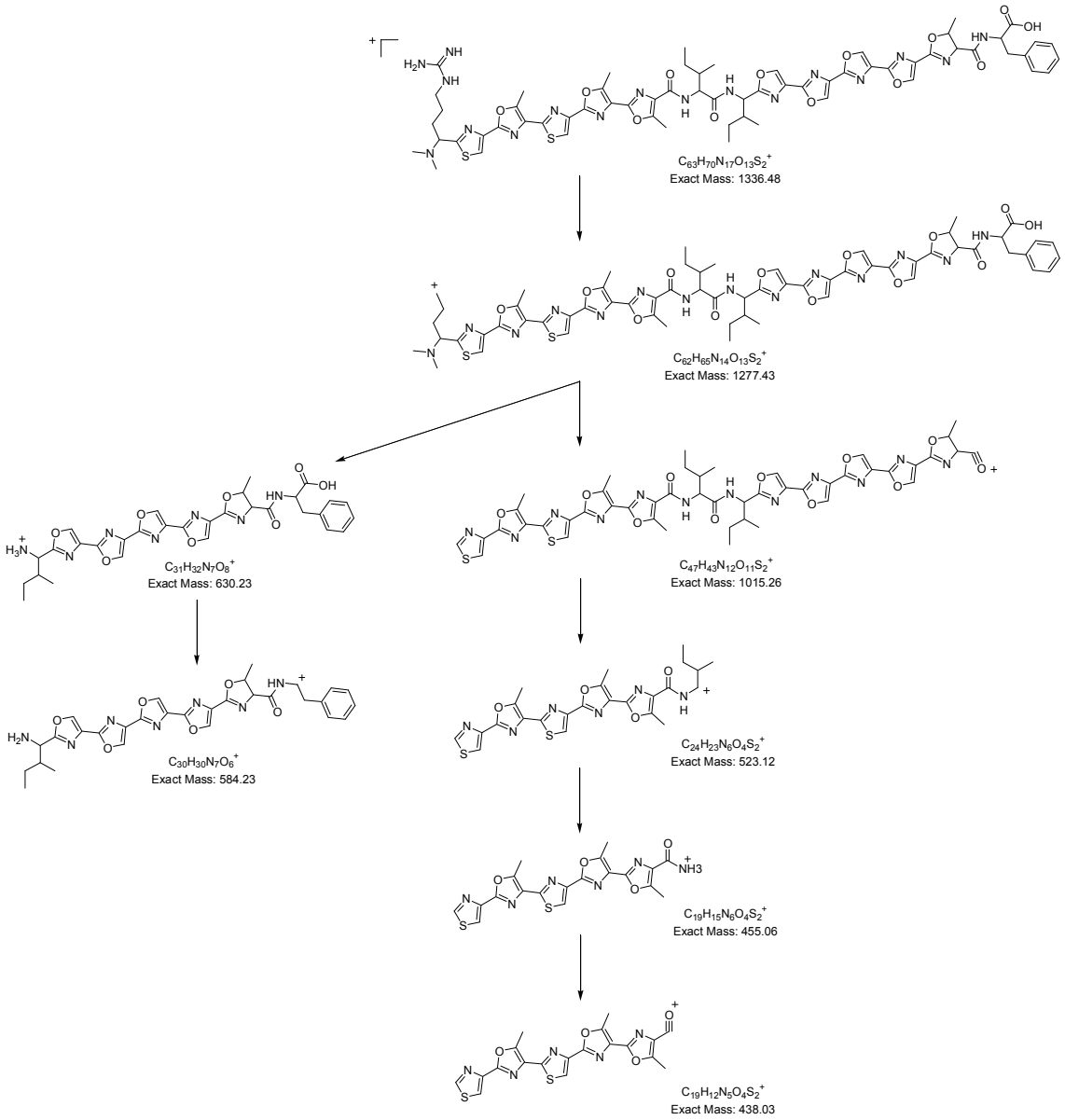

Figure S2. MS/MS-fragmentation scheme and peak assignment of plantazolicin A (**1a**). Fragmentation at the *N*-terminal side of Plantazolicin A.

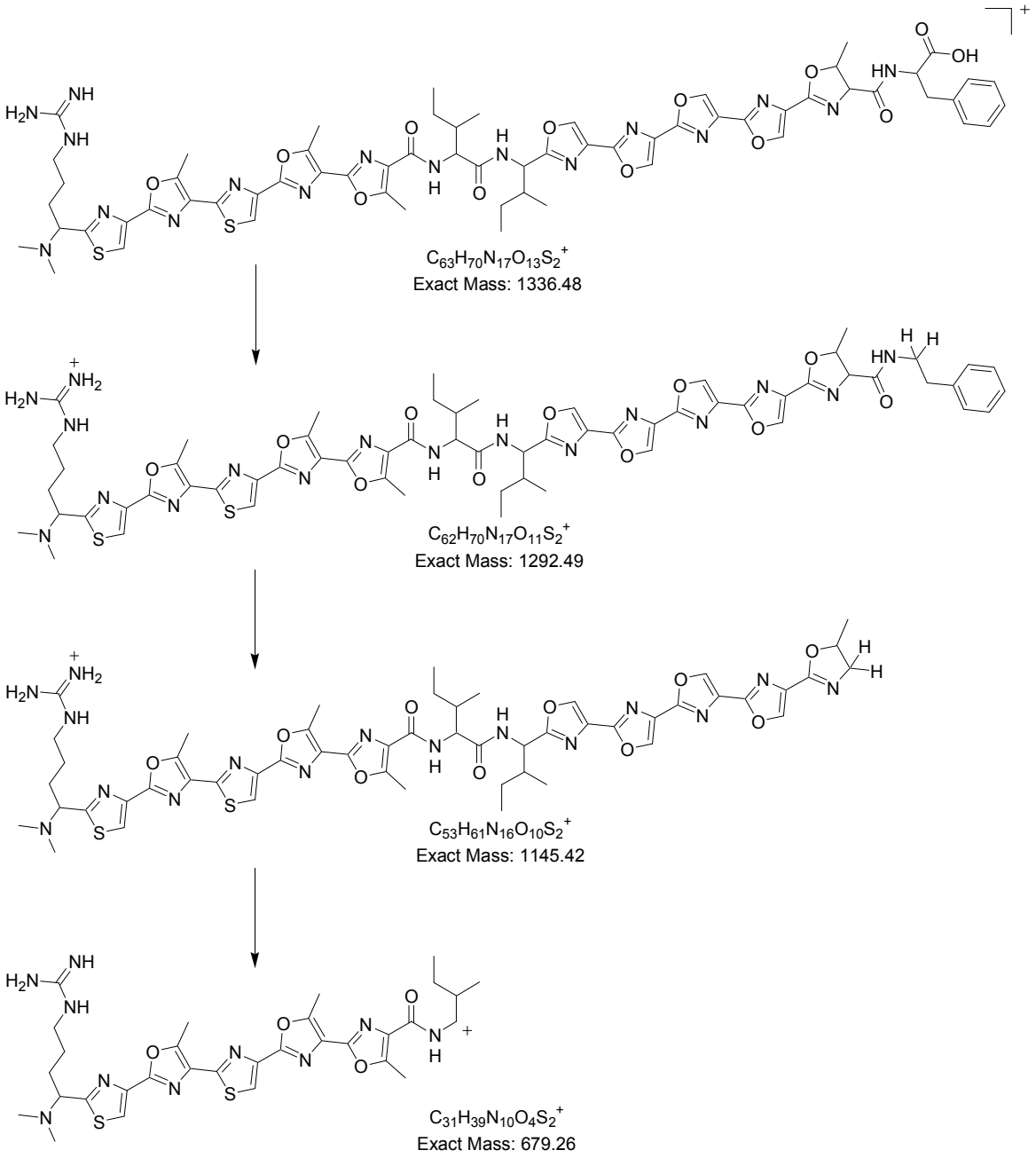

Figure S2. MS/MS-fragmentation scheme and peak assignment of plantazolicin A (**1a**) (continued). Fragmentation at the *C*-terminal side of Plantazolicin A.

Table S2. NMR spectroscopic data of plantazolicin A in DMSO-*d6*

	Position	***δ (^{1}H) (p.p.m.) J in Hz***	***δ (^{13}C) (p.p.m.)***	***δ (^{15}N) (p.p.m.)***
N,*N*-diMeArg[1]	α	3.93 (t, 6.10)	65.2	30.7
	β	1.52 (m)	24.2	—
	γ	0.87, 1.24 (m)	22.2	—
	δ	1.85, 1.98 (m)	36.9	—
	ε	9.23	—	84.86
	ζ	—	n.d.	—
	η^1/η^2	7.67, 7.72 (m)	—	109.17, 123.62
	gem-Me	2.26 (s)	41.1	—
Thz[2]	2	—	172.6	—
	3	—	—	312.1
	4	—	141.0	—
	5	8.41 (s)	122.2	—
5-MeOxz[3]	2	—	173.3	—
	3	—	—	250.0
	4	—	129.7	—
	5	—	147.4	—
	5-CH_3	2.83 (s)	11.4	—
Thz[4]	2	—	161.5	—
	3	—	—	303.7
	4	—	142.5	—
	5	8.47 (s)	121.5	—
5-MeOxz[5]	2	—	n.d.	—
	3	—	—	249.5
	4	—	124.8	—
	5	—	150.5	—
	5-CH_3	2.75 (s)	11.3	—
5-MeOxz[6]	2	—	n.d.	—
	3	—	—	246.9
	4	—	129.0	—
	5	—	152.2	—
	5-CH_3	2.64 (s)	11.00	—
	C(O)	—	n.d.	—
Ile[7]	NH	7.89 (d, 9.2)	—	113.9
	α	4.48 (t, 8.2)	56.4	—
	β	1.93 (m)	36.7	—
	γ1	1.05 (d, 6.6)	21.5	—
	γ2	0.87 (m)	22.3	—
	δ	0.85 (m)	10.6	—
	C(O)	—	176.9	—
Ile[8]	NH	8.78 (d, 7.1)	—	118.6
	α	4.93 (t, 7.8)	51.2	—
	β	2.07 (m)	36.6	—
	γ1	1.05 (d, 6.6)	21.5	—
	γ2	0.84	22.6	—
	δ	0.85 (m)	10.6	—
Oxz[9]	2	—	164.2	—

	3	—	—	248.5
	4	—	n.d.	—
	5	8.96 (s)	140.6	—
Oxz[10]	2	—	155.2	—
	3	—	—	246.8
	4	—	n.d.	—
	5	9.08 (s)	140.6	—
Oxz[11]	2	—	n.d.	—
	3	—	—	247.2
	4	—	n.d.	—
	5	9.11 (s)	140.8	—
Oxz[12]	2	—	154.7	—
	3	—	—	250.1
	4	—	n.d.	—
	5	8.81 (s)	142.5	—
5-MeOxl[13]	2	—	156.9	—
	3	—	—	222.1
	4	4.23 (d, 7.6)	74.3	—
	5	4.61 (m)	79.4	—
	5-CH_3	1.44 (d, 6.2)	20.9	—
	C(O)	—	168.53	—
Phe[14]	NH	7.23 (d, 6.3)	—	122.0
	α	4.10 (t, 5.3)	54.4	—
	β	2.92 (d, 12.3), 3.03 (d, 5.8)	37.1	—
	γ	—	138.4	—
	δ1/δ2	6.96 (d, 7.4)	129.15	—
	ε1/ε2	7.02 (t, 7.1)	127.1	—
	ζ	6.99 (d, 6.9)	125.3	—
	C(O)	—	n.d.	—

n.d. = not detected
N,*N*-diMeArg = *N*,*N*-dimethylarginine, Thz = thiazole, 5-MeOxz = 5-methyloxazole, Ile = isoleucine, Oxz = oxazole, 5-MeOxl = 5-methyloxazolidine and Phe[14] = phenylalanine.

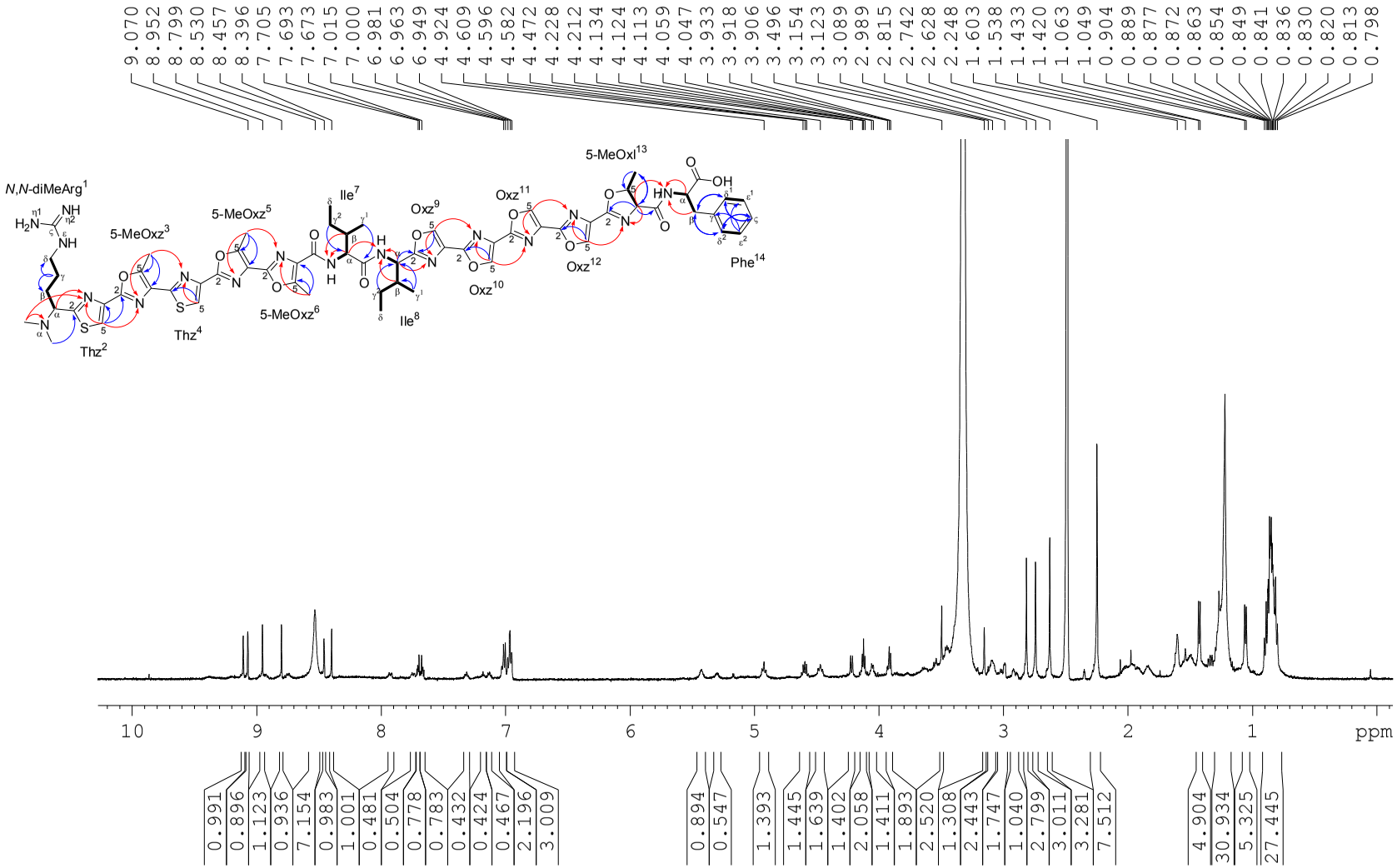

Figure S3. ^{1}H-NMR spectrum of plantazolicin A (**1a**) in DMSO-*d6*

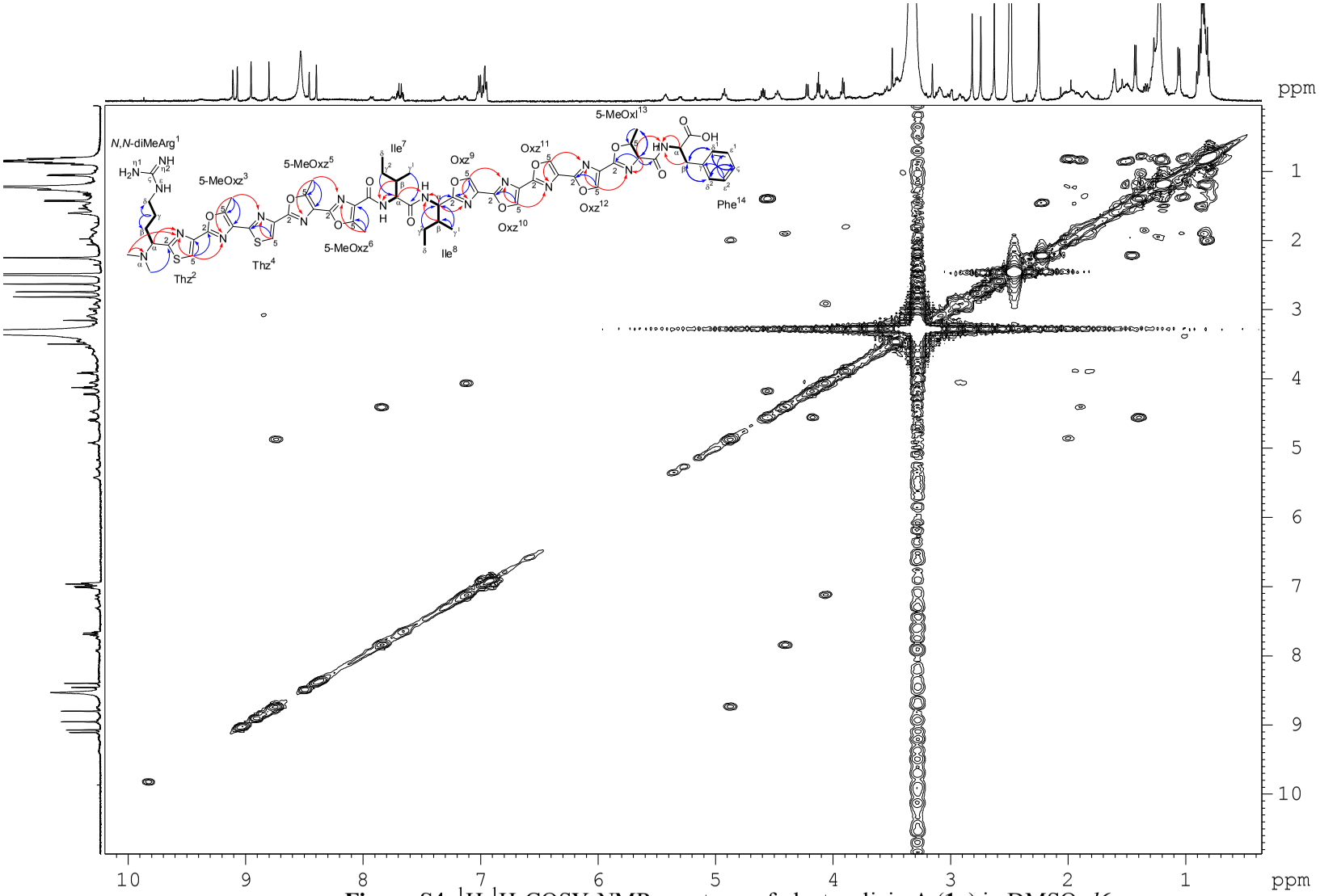

Figure S4. ^{1}H-^{1}H-COSY-NMR spectrum of plantazolicin A (**1a**) in DMSO-*d6*

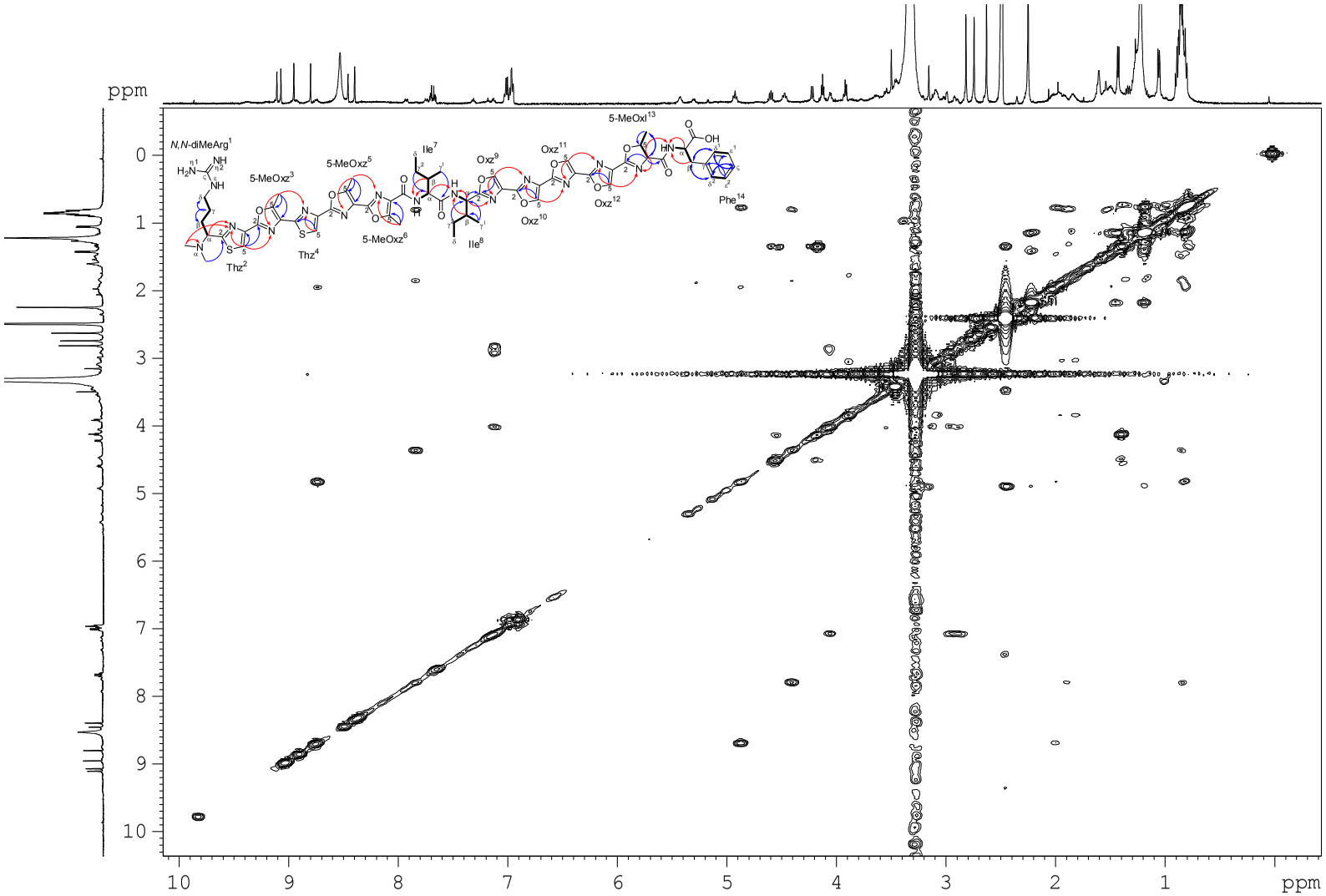

Figure S5. ^{1}H-^{1}H-TOCSY-NMR spectrum of plantazolicin A (**1a**) in DMSO-*d6*

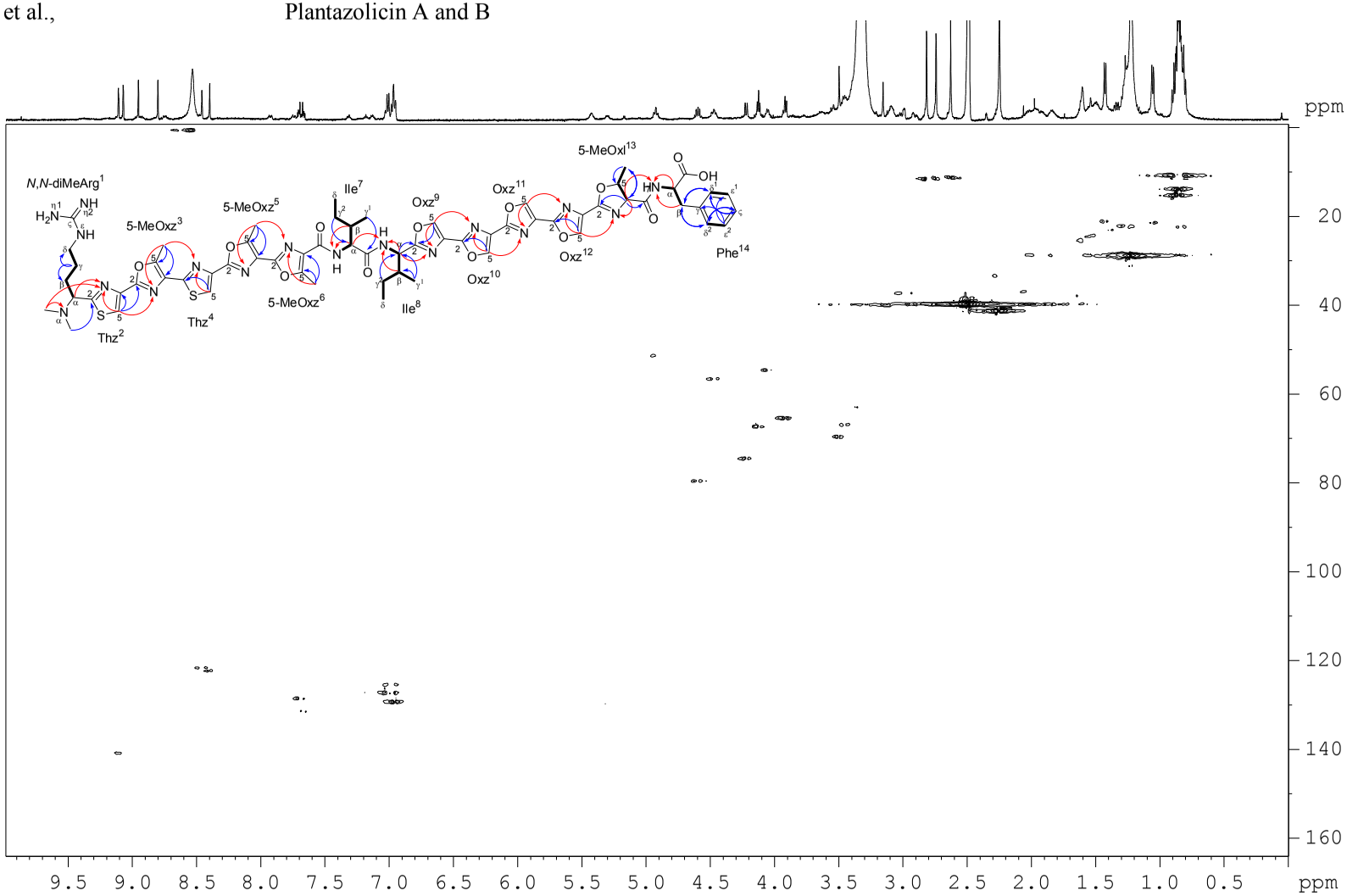

Figure S6. ^{1}H-^{13}C-HSQC-NMR spectrum of plantazolicin A (**1a**) in DMSO-*d6*

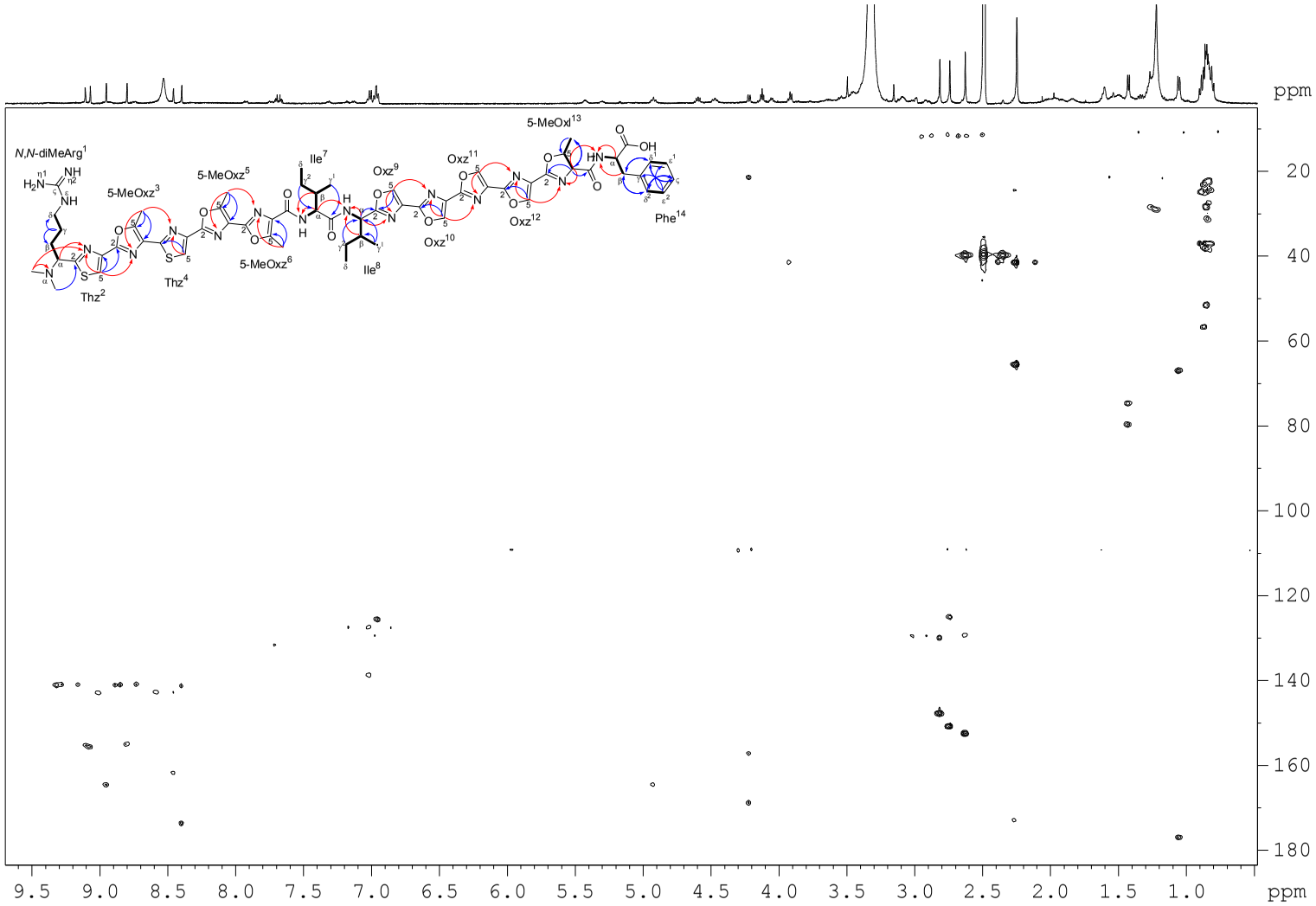

Figure S7. ^{1}H-^{13}C-HMBC-NMR spectrum of plantazolicin A (**1a**) in DMSO-*d6*

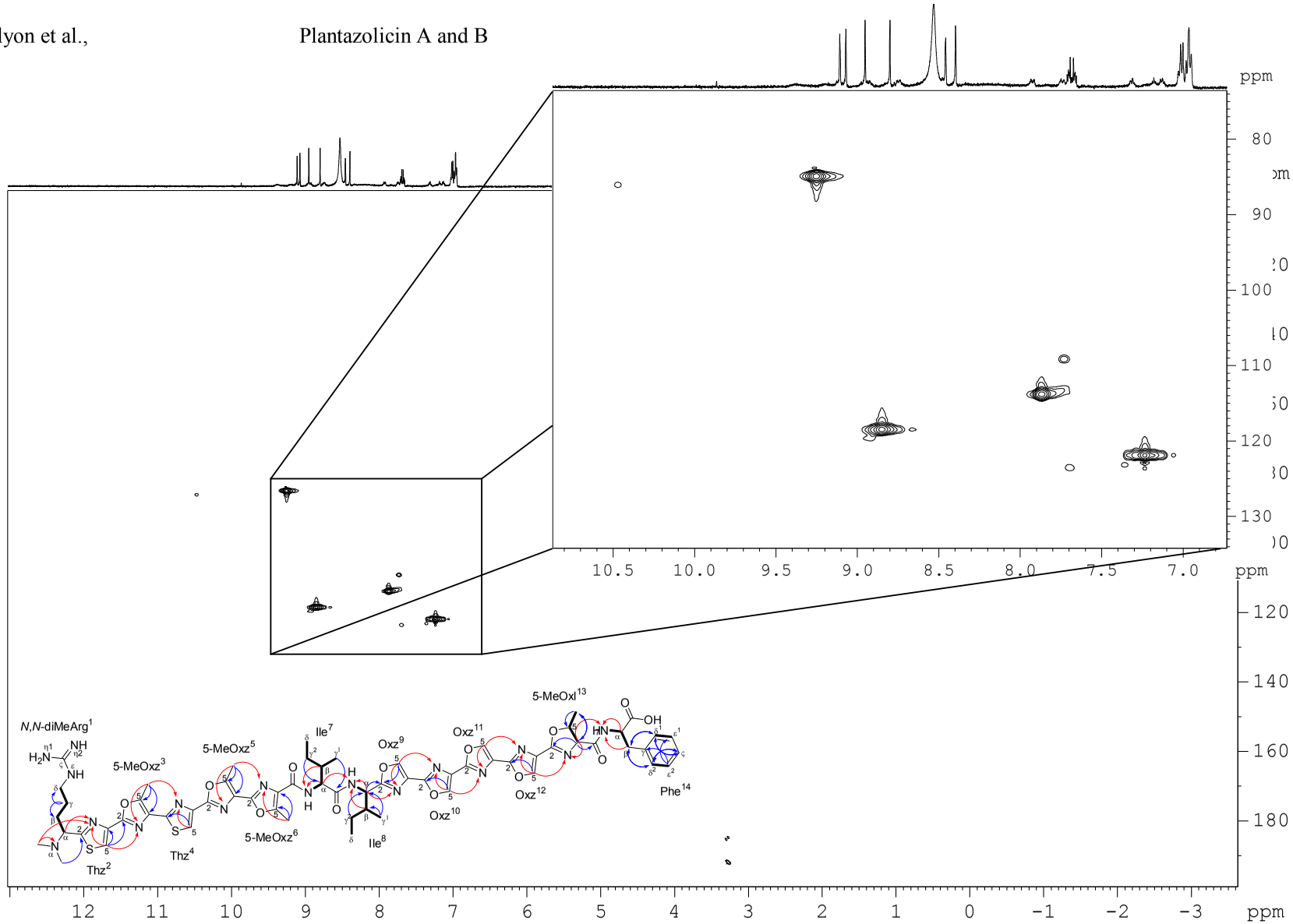

Figure S8. ^{1}H-^{15}N-HMQC-NMR spectrum of plantazolicin A (**1b**) in DMSO-*d6*

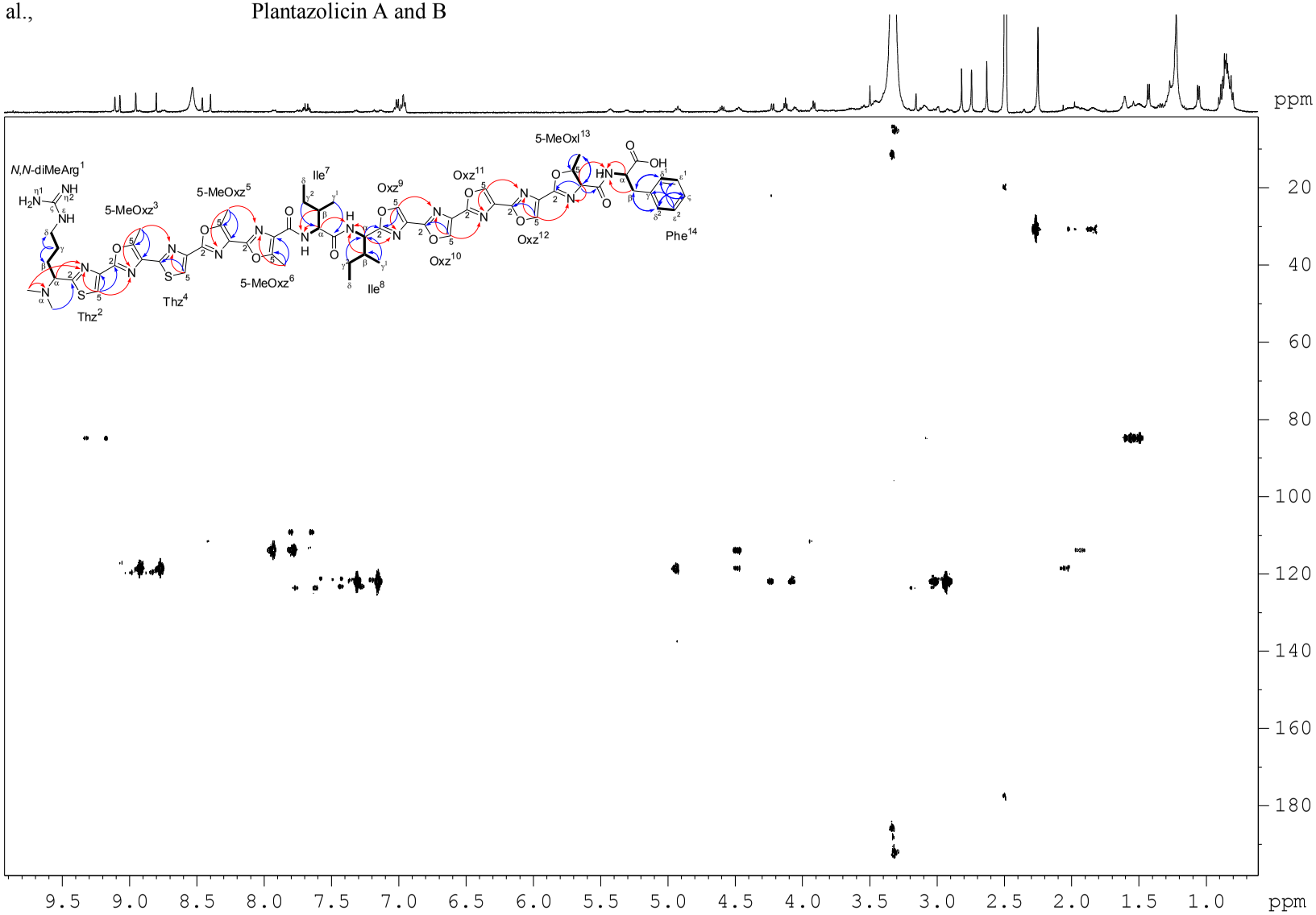

Figure S9. ^{1}H-^{15}N-HMBC-NMR spectrum of plantazolicin A (**1b**) in DMSO-*d6*, on 600 MHz center at 100 ppm.

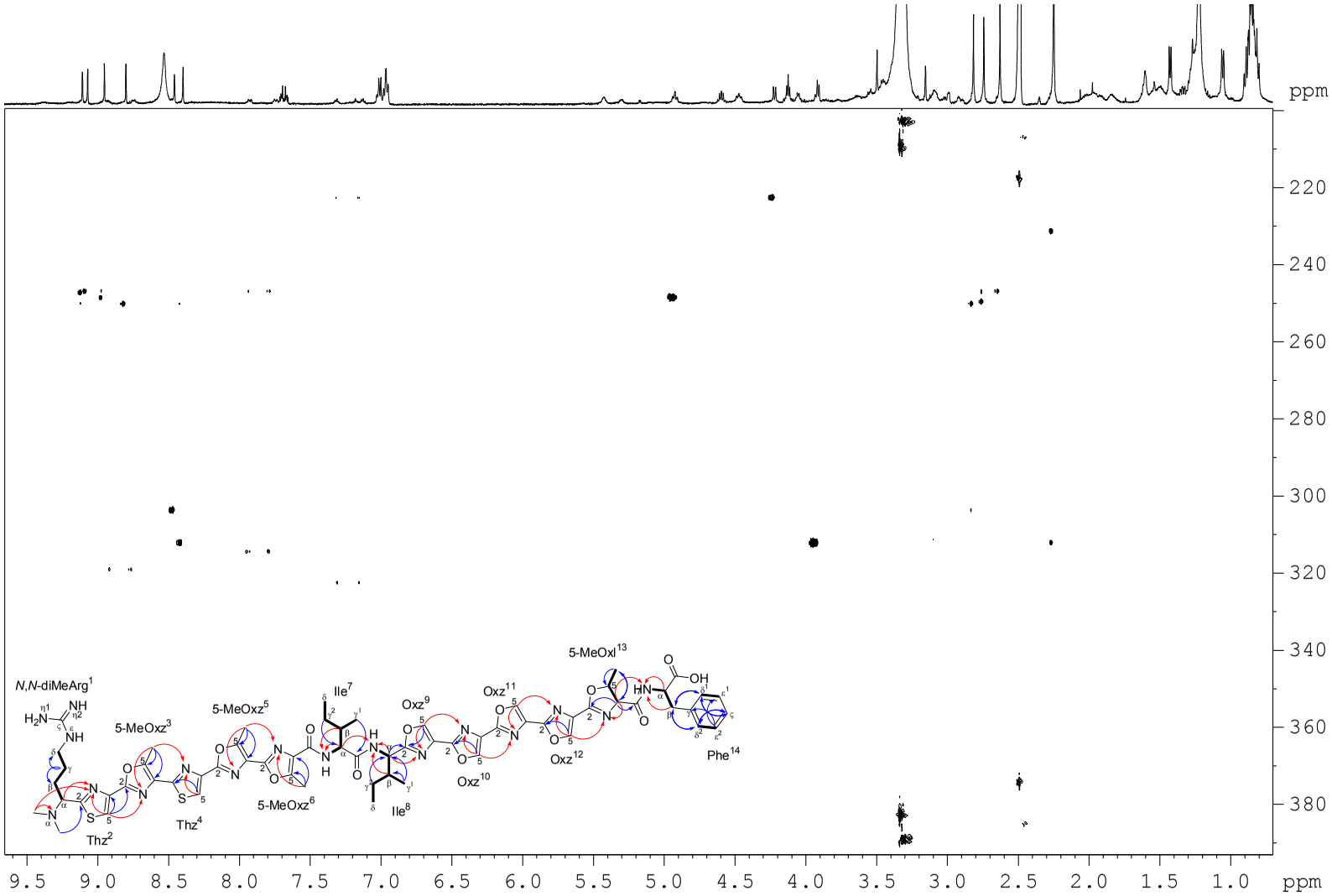

Figure S10. ^{1}H-^{15}N-HMBC-NMR spectrum of plantazolicin A (**1b**) in DMSO-*d6*, on 600 MHz center at 300 ppm.

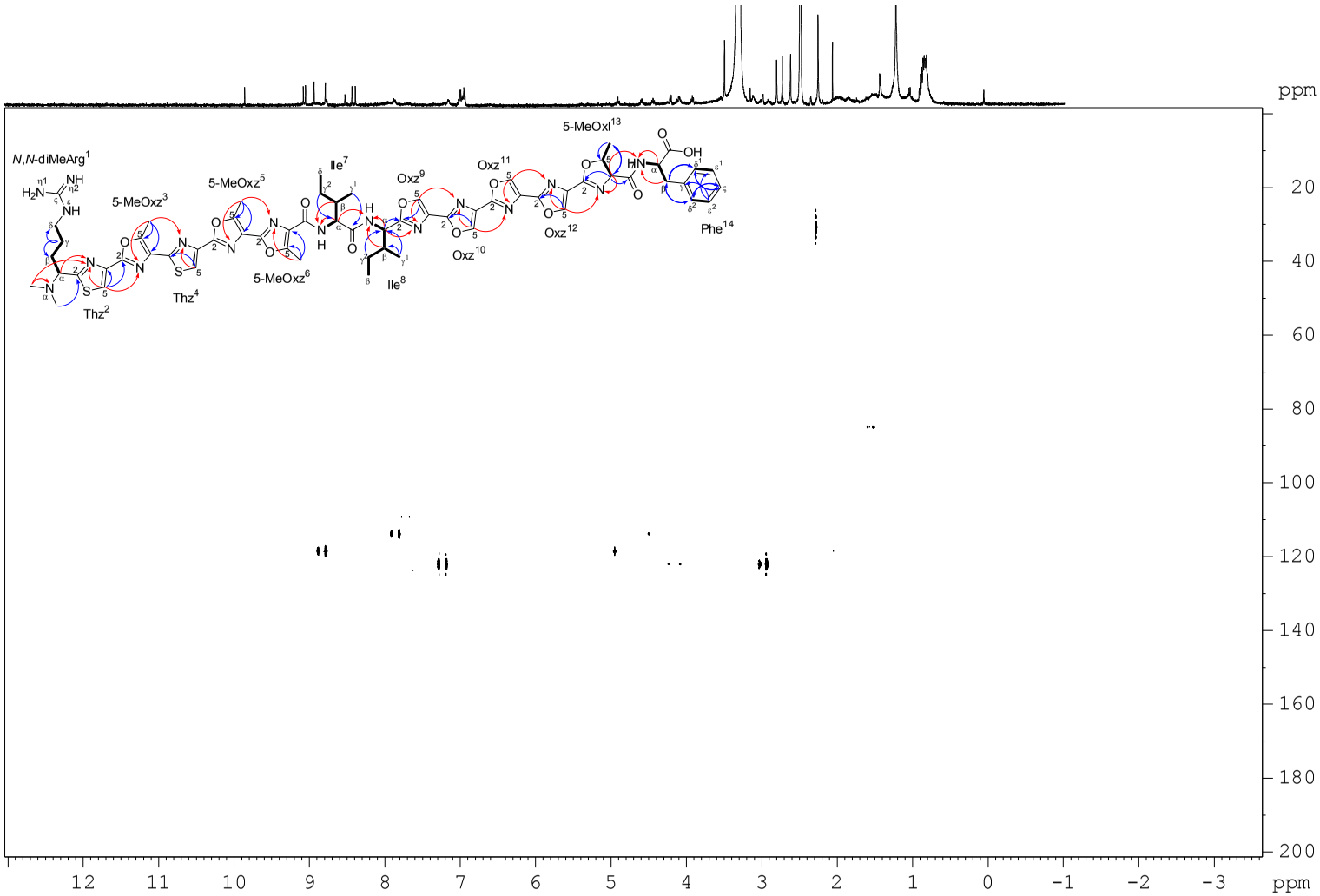

Figure S11. ^{1}H-^{15}N-HMBC-NMR spectrum of plantazolicin A (**1b**) in DMSO-*d6*, on 900 MHz center at 100 ppm.

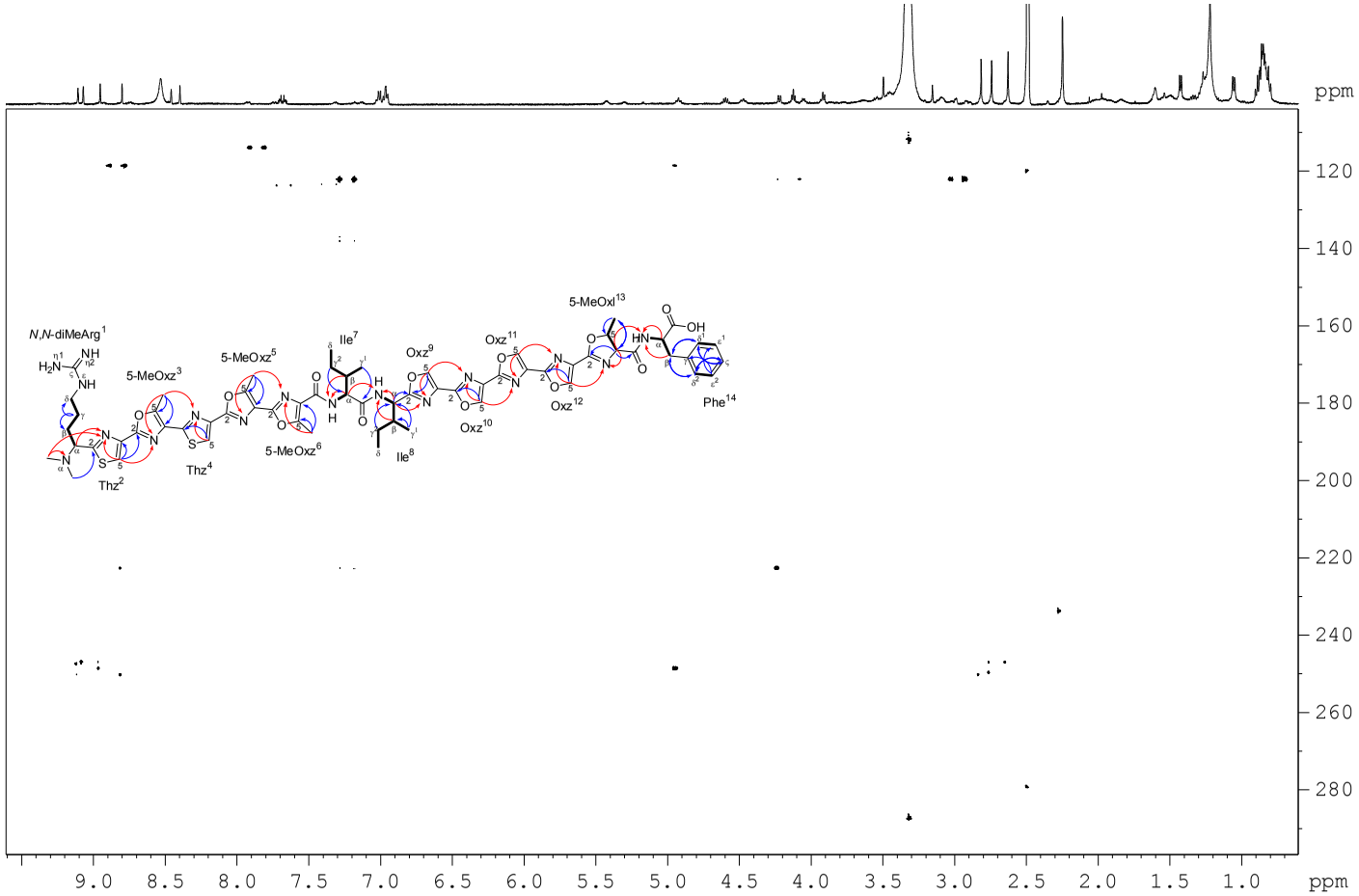

Figure S12. ^{1}H-^{15}N-HMBC-NMR spectrum of plantazolicin A (**1b**) in DMSO-*d6*, on 900 MHz center at 200 ppm.

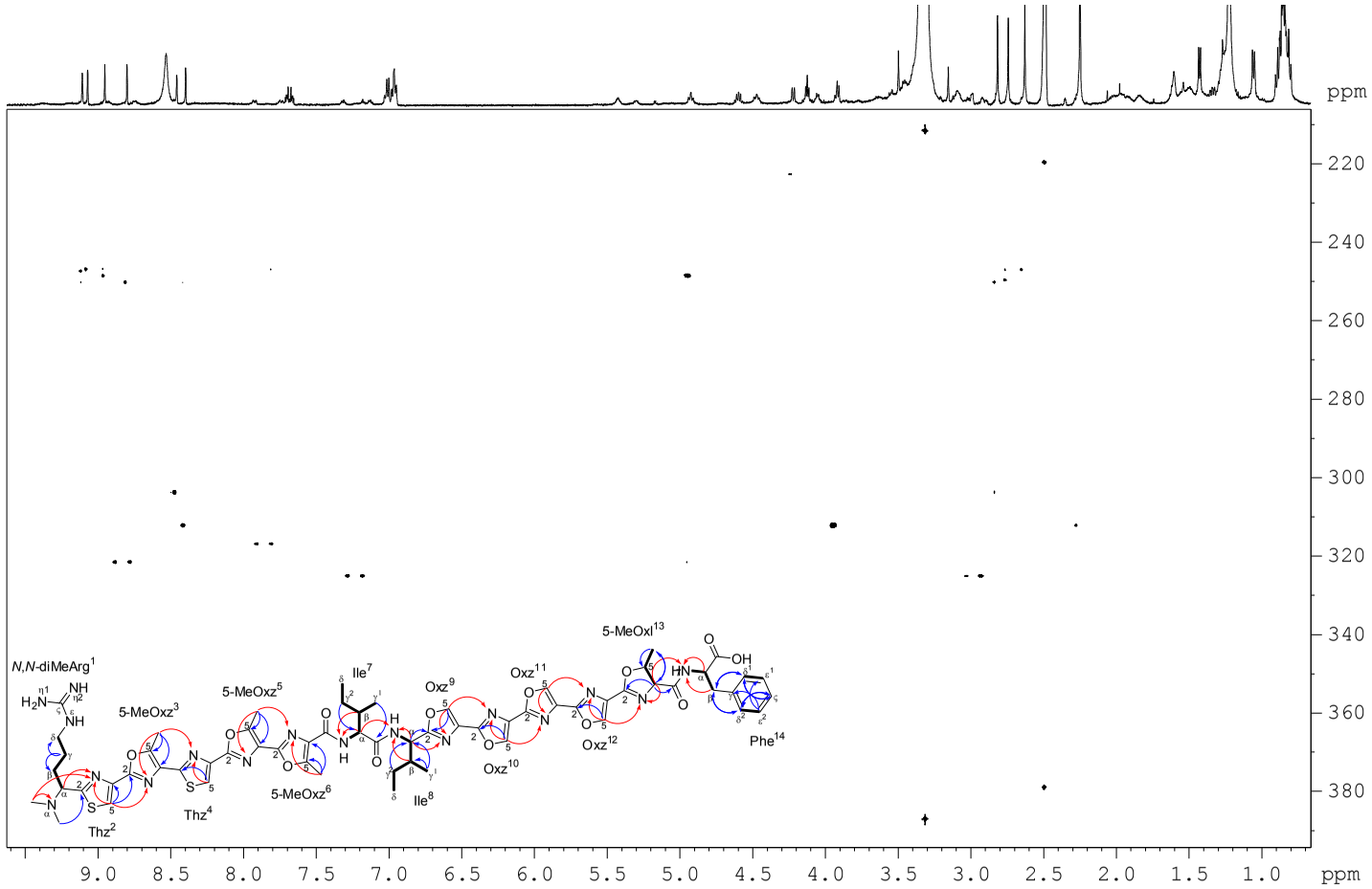

Figure S13. ^{1}H-^{15}N-HMBC-NMR spectrum of plantazolicin A (**1b**) in DMSO-*d6*, on 900 MHz center at 300 ppm.

References

[1] Spizizen, J. *Proc. Natl. Acad. Sci.* **1958**, *44*, 1072-1078.

[2] Scholz, R.; Molohon, K. J.; Nachtigall, J.; Vater, J.; Markley, A. L.; Süssmuth, R. D.; Mitchell, D. A.; Borriss, R. *J. Bacteriol.* **2011**, *193*, 215-224.