Org. Biomol. Chem.

Oxoanion binding to a cyclic pseudopeptide containing 1,4-disubstituted 1,2,3-triazole moieties

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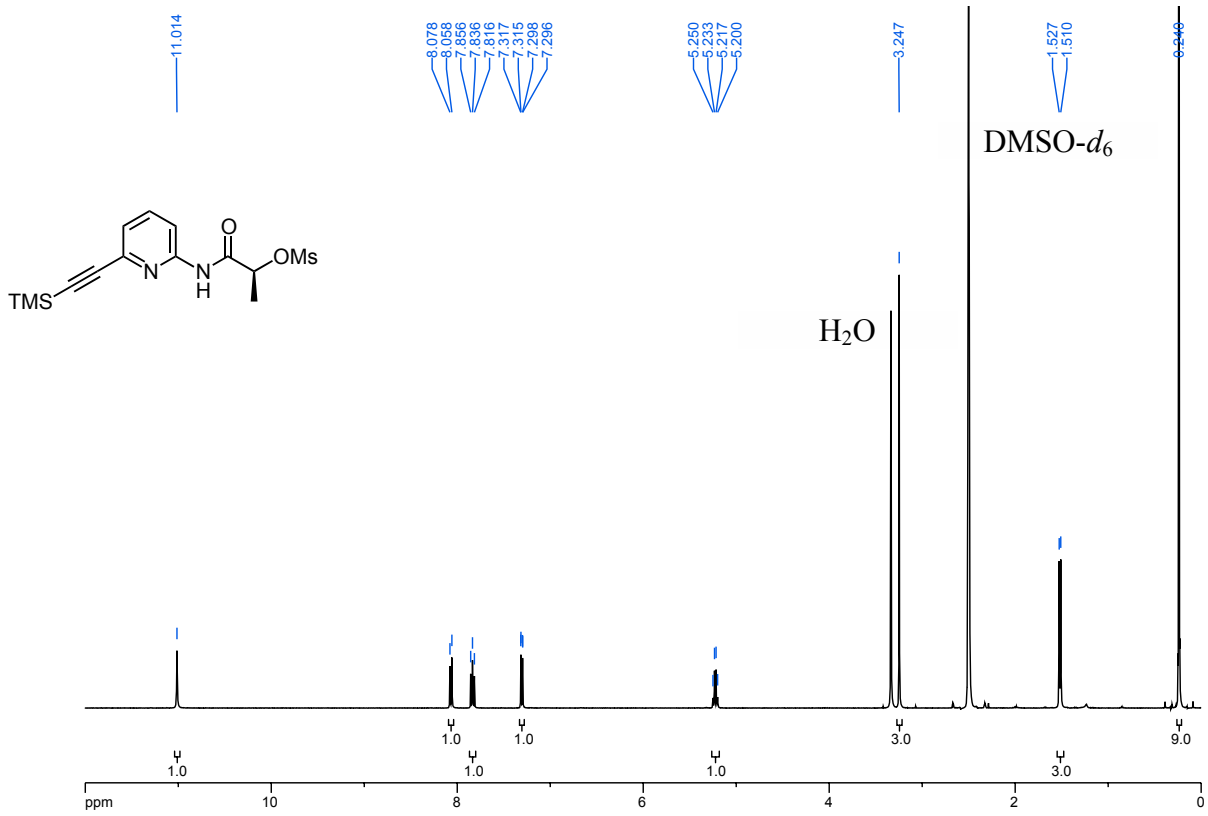
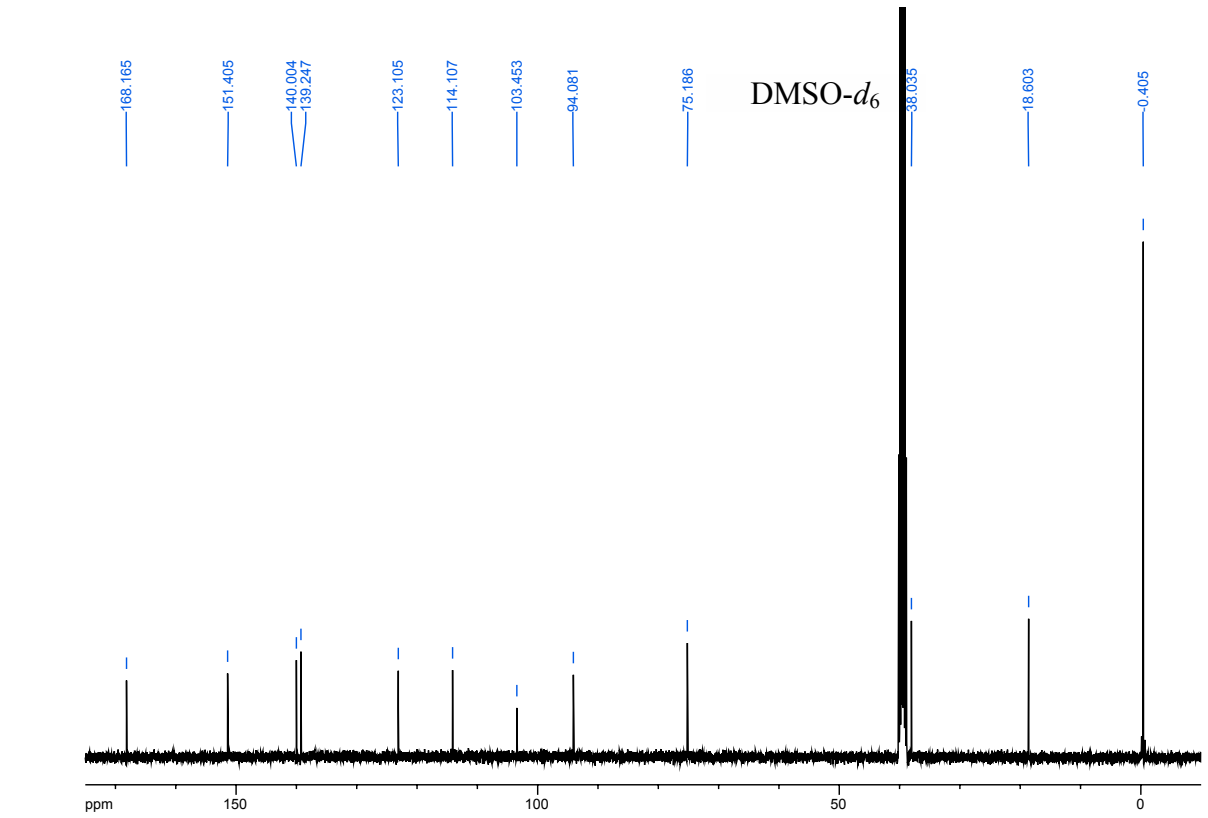
CONTENT

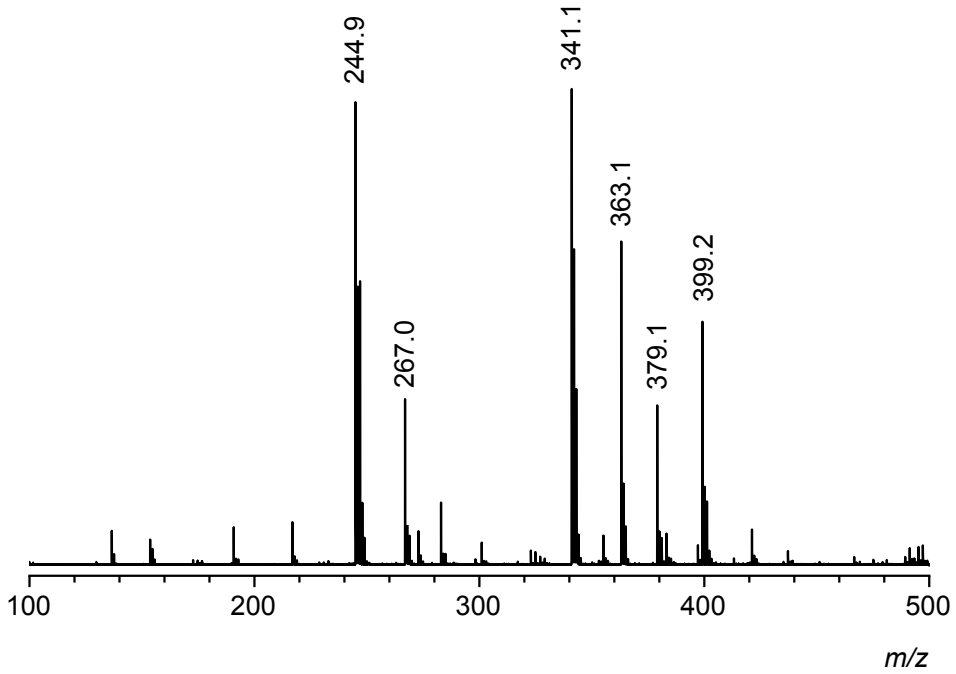
^{1}H NMR: TMS-Epa-(*S*)-Lac-OMs **5** (400 MHz, DMSO-d_6).



^{13}C NMR: TMS-Epa-(*S*)-Lac-OMs **5** (101 MHz, DMSO-d_6).

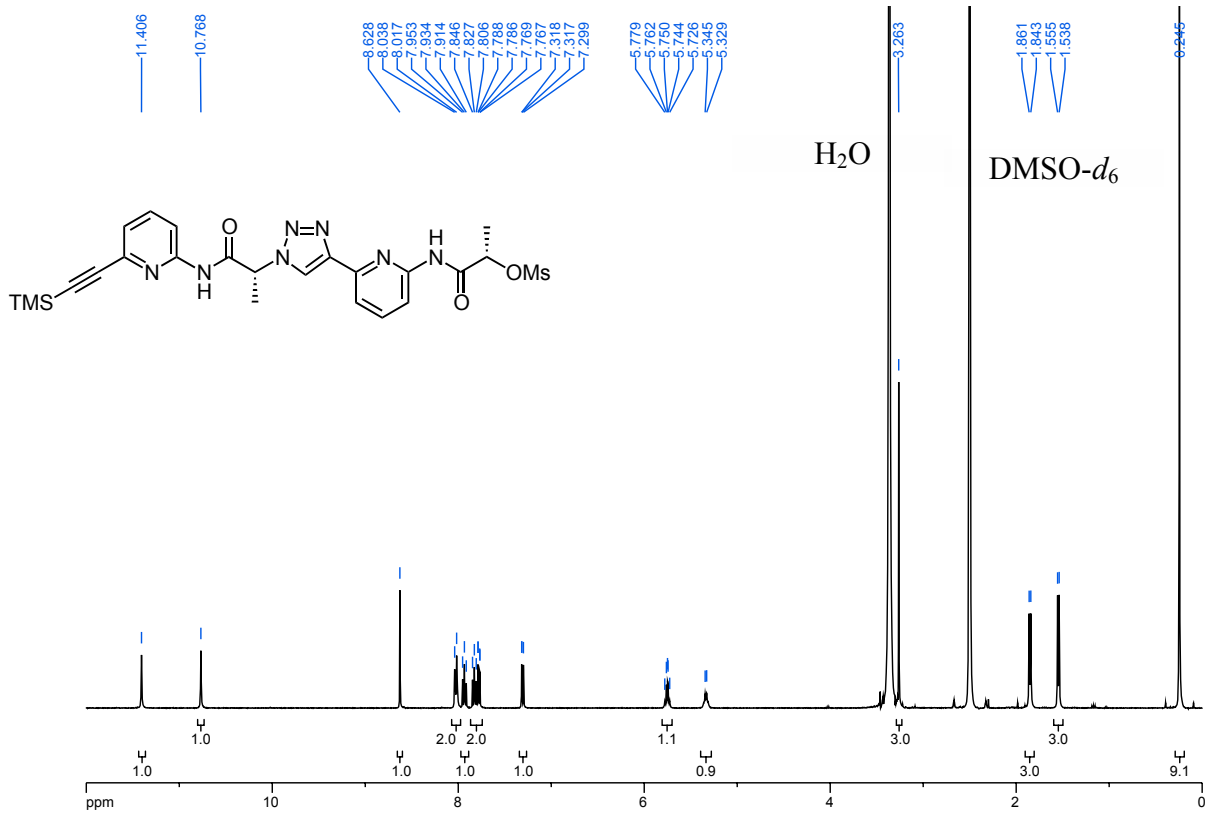


MALDI-TOF MS: TMS-Epa-(*S*)-Lac-OMs **5** (positive mode).

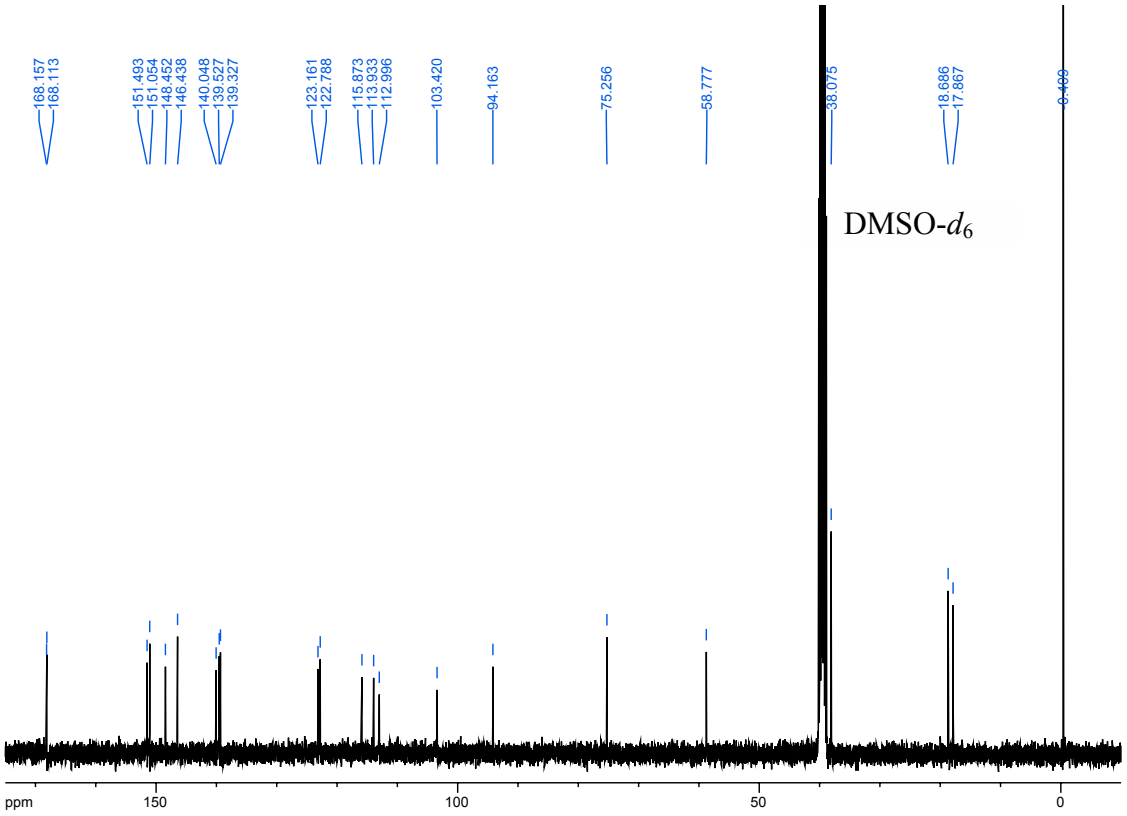


		m/z calcd.	*m/z exp.*
$[M–CH_3SO_3H+H]^+$	$C_{14}H_{20}N_2O_4SSi – CH_3SO_3H + H^+$	245.1	244.9
$[M–CH_3SO_3H+Na]^+$	$C_{14}H_{20}N_2O_4SSi – CH_3SO_3H + Na^+$	267.1	267.0
$[M+H]^+$	$C_{14}H_{20}N_2O_4SSi + H^+$	341.1	341.1
$[M+Na]^+$	$C_{14}H_{20}N_2O_4SSi + Na^+$	363.1	363.1
$[M+K]^+$	$C_{14}H_{20}N_2O_4SSi + K^+$	379.1	379.1
$[M+C_3H_6O+H]^+$	$C_{14}H_{20}N_2O_4SSi + C_3H_6O + H^+$	399.1	399.2

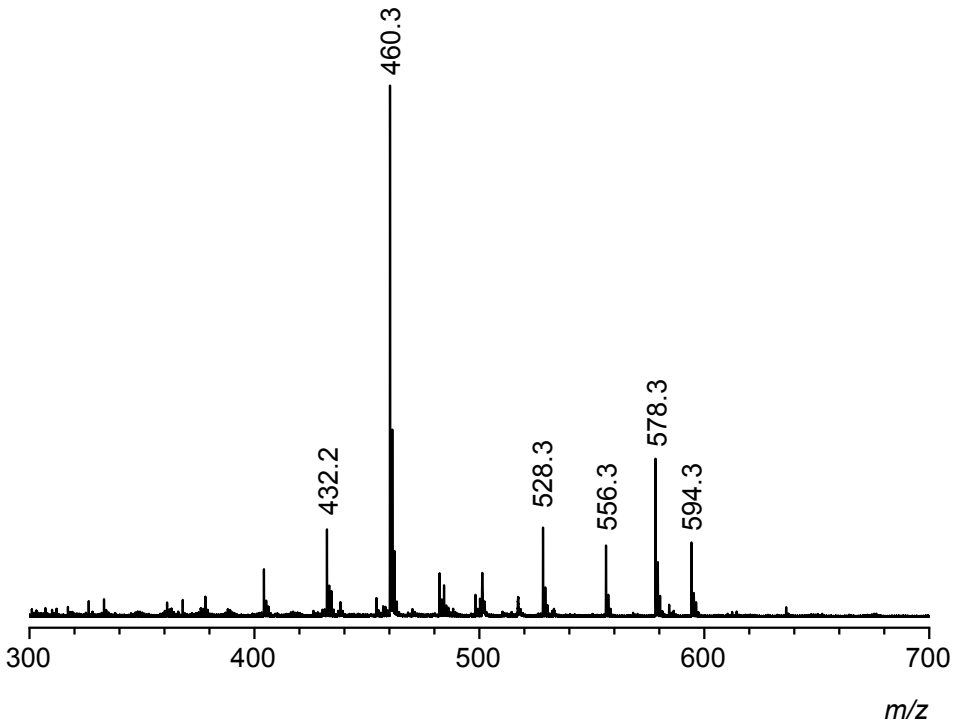
^{1}H NMR: TMS-Epa-(*R*)-Lac-1,4-Tri-Epa-(*S*)-Lac-OMs **7** (400 MHz, DMSO-d_6).



^{13}C NMR: TMS-Epa-(*R*)-Lac-1,4-Tri-Epa-(*S*)-Lac-OMs **7** (101 MHz, DMSO-d_6).

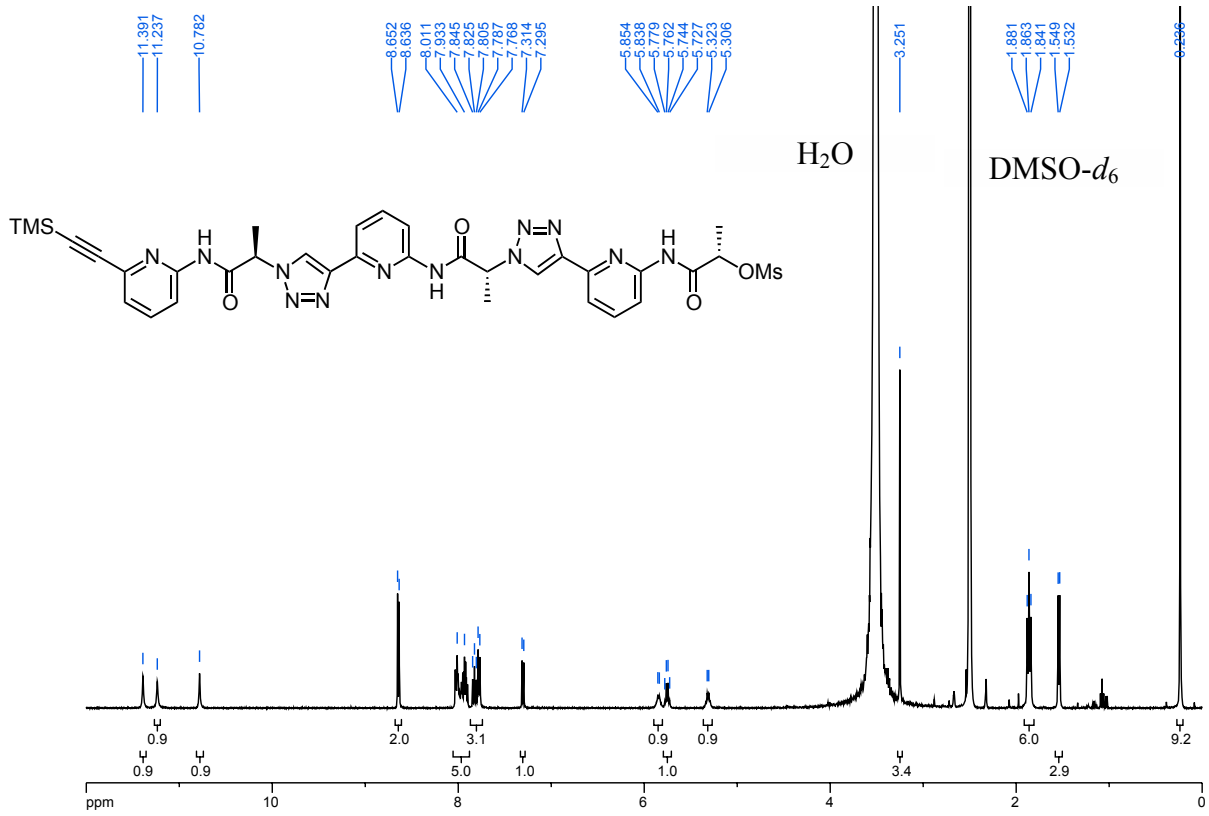


MALDI-TOF MS: TMS-Epa-(*R*)-Lac-1,4-Tri-Epa-(*S*)-Lac-OMs **7** (positive mode).

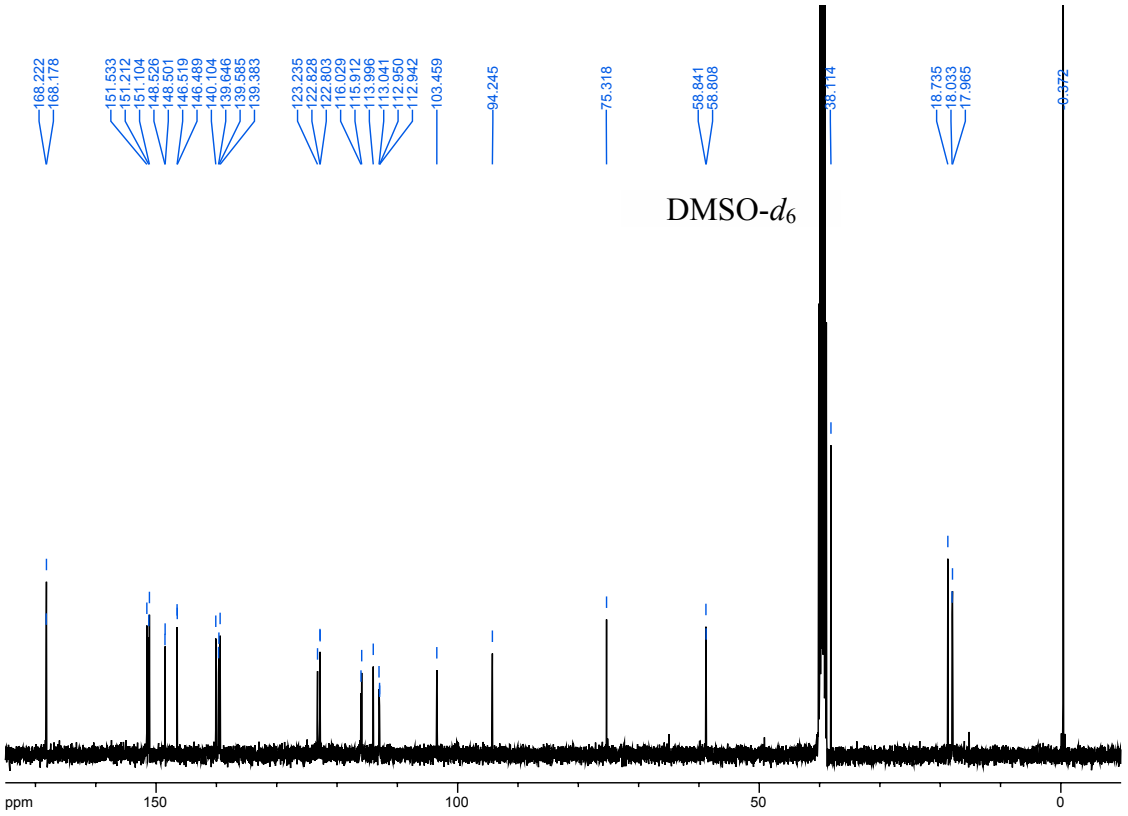


		m/z calcd.	*m/z exp.*
$[M–N_2–CH_3SO_3H+H]^+$	$C_{24}H_{29}N_7O_5SSi – N_2 – CH_3SO_3H + H^+$	432.6	432.2
$[M–CH_3SO_3H+H]^+$	$C_{24}H_{29}N_7O_5SSi – CH_3SO_3H + H^+$	460.6	460.3
$[M–N_2+H]^+$	$C_{24}H_{29}N_7O_5SSi – N_2 + H^+$	528.7	528.3
$[M+H]^+$	$C_{24}H_{29}N_7O_5SSi + H^+$	556.2	556.3
$[M+Na]^+$	$C_{24}H_{29}N_7O_5SSi + Na^+$	578.2	578.3
$[M+K]^+$	$C_{24}H_{29}N_7O_5SSi + K^+$	594.1	594.3

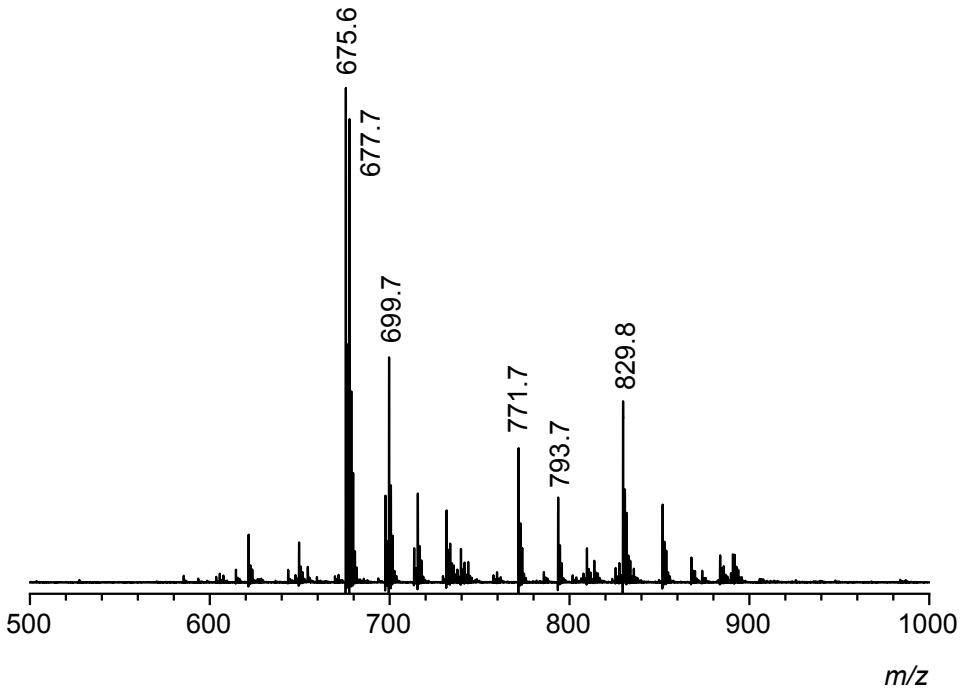
^{1}H NMR: TMS-Epa-[(*R*)-Lac-1,4-Tri-Epa]$_2$-(*S*)-Lac-OMs **8** (400 MHz, DMSO-d_6).



^{13}C NMR: TMS-Epa-[(*R*)-Lac-1,4-Tri-Epa]$_2$-(*S*)-Lac-OMs **8** (101 MHz, DMSO-d_6).

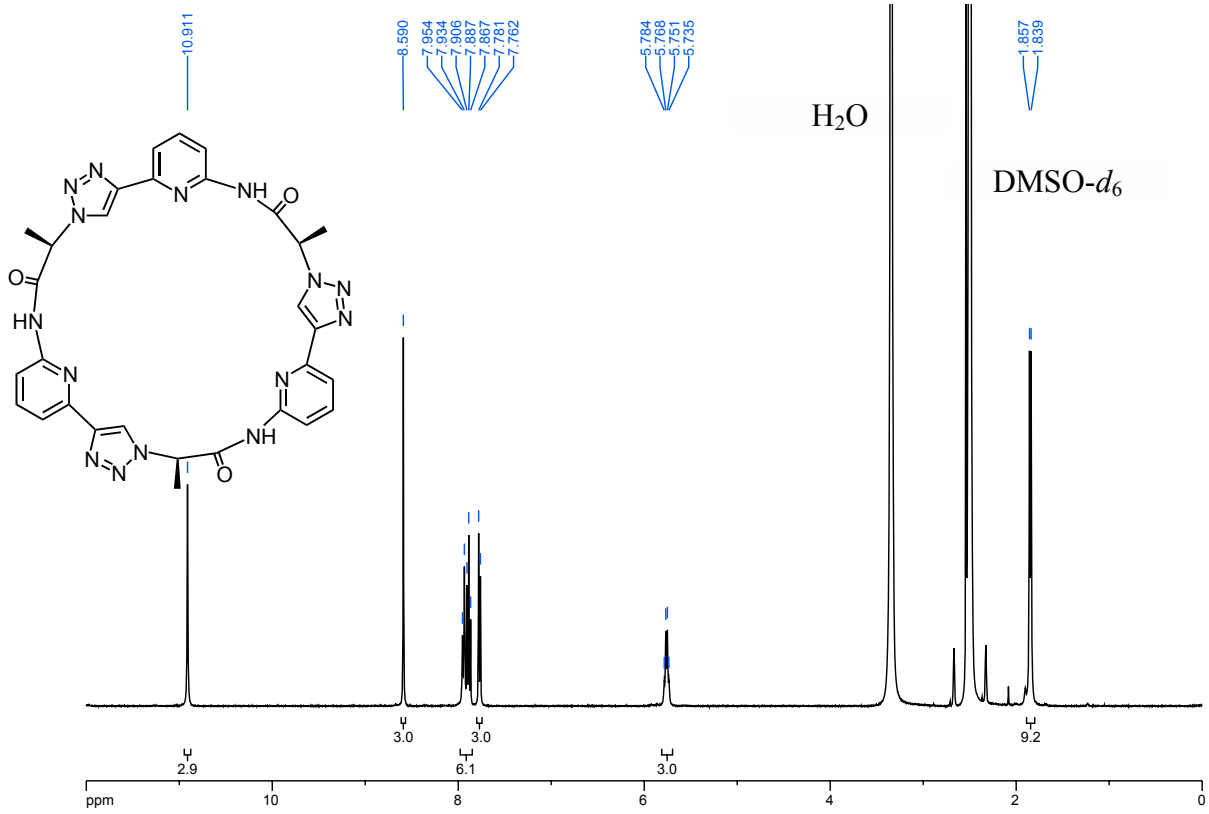


MALDI-TOF MS: TMS-Epa-[(*R*)-Lac-1,4-Tri-Epa]$_2$-(*S*)-Lac-OMs **8** (positive mode).

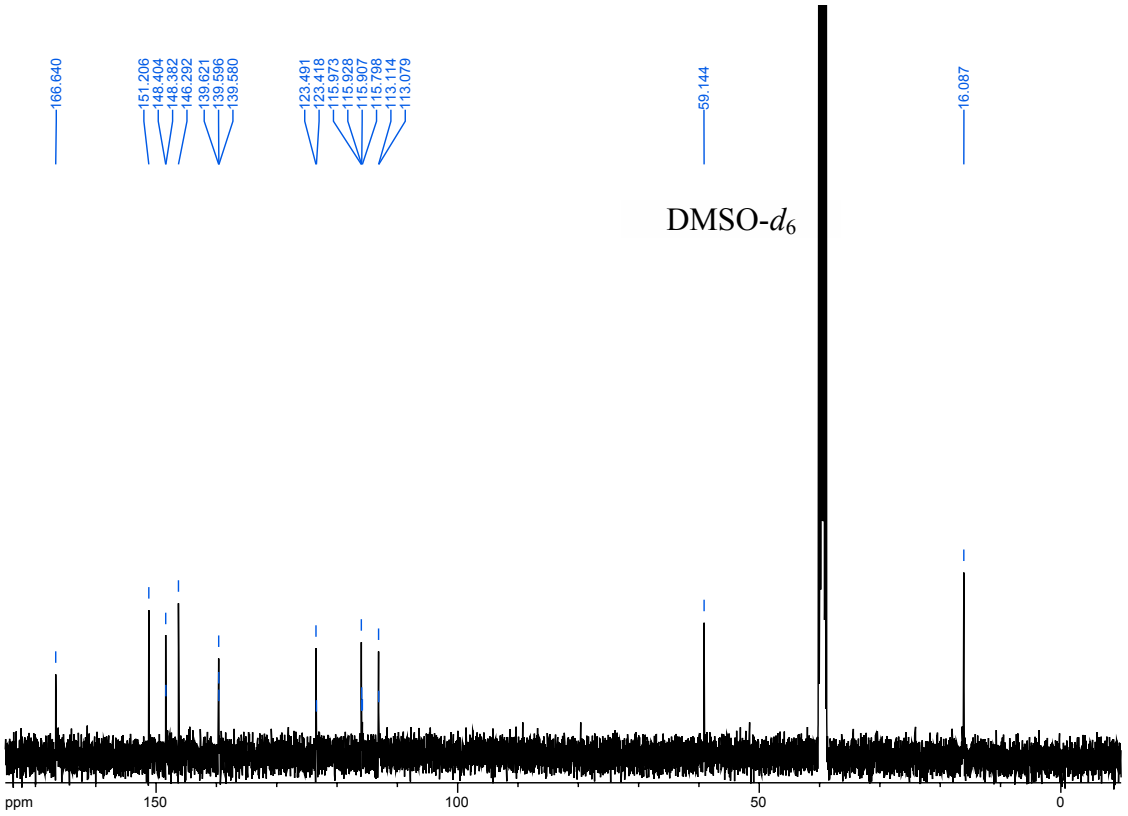


		m/z calcd.	*m/z exp.*
$[M–CH_3SO_3H+H]^+$	$C_{34}H_{38}N_{12}O_6SSi – CH_3SO_3H + H^+$	675.8	675.6
$[M–CH_3SO_3H+H_2+H]^+$	$C_{34}H_{38}N_{12}O_6SSi – CH_3SO_3H + H_2 + H^+$	677.8	677.7
$[M–CH_3SO_3H+H_2+Na]^+$	$C_{34}H_{38}N_{12}O_6SSi – CH_3SO_3H + H_2 + Na^+$	699.8	699.7
$[M+H]^+$	$C_{34}H_{38}N_{12}O_6SSi + H^+$	771.3	771.7
$[M+Na]^+$	$C_{34}H_{38}N_{12}O_6SSi + Na^+$	793.2	793.7
$[M+C_3H_6O+H]^+$	$C_{34}H_{38}N_{12}O_6SSi + C_3H_6O + H^+$	829.3	829.8

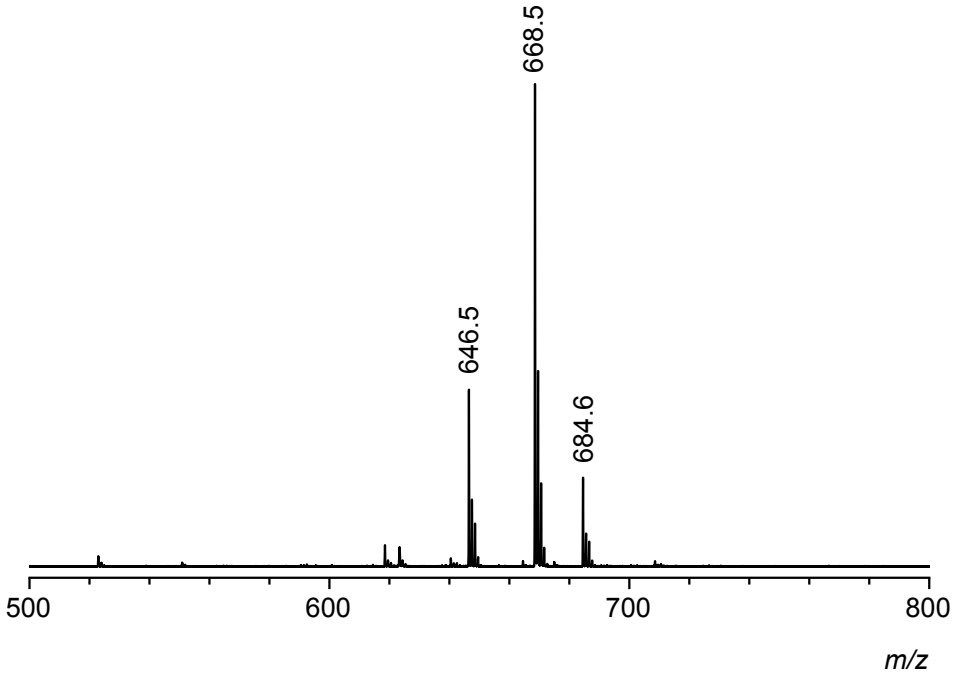
^{1}H NMR: *cyclo*[(*R*)-Lac-1,4-Tri-Epa]$_3$ **3** (400 MHz, DMSO-d_6).



^{13}C NMR: *cyclo*[(*R*)-Lac-1,4-Tri-Epa]$_3$ **3** (101 MHz, DMSO-d_6).

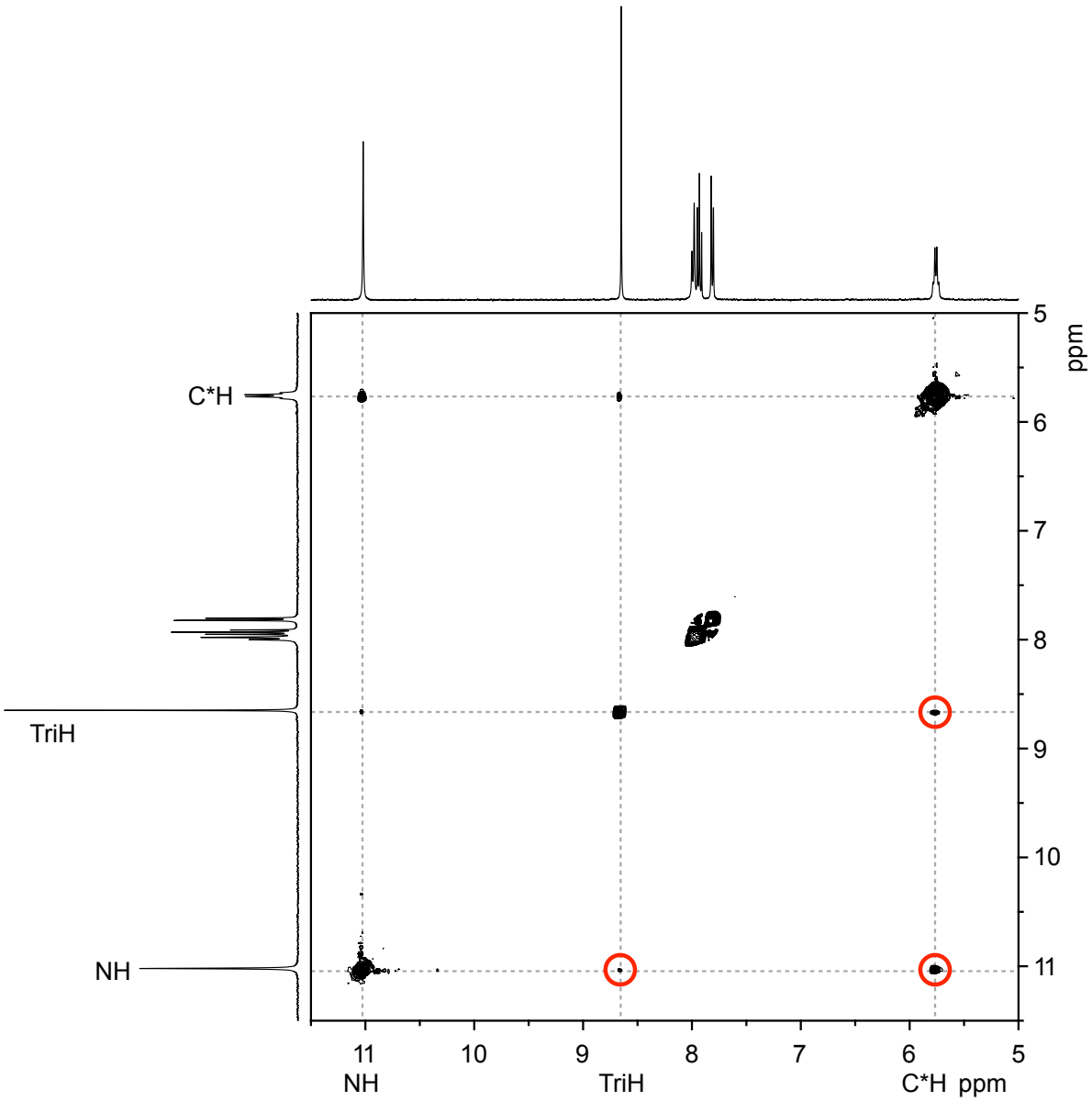


MALDI-TOF MS: *cyclo*[(*R*)-Lac-1,4-Tri-Epa]$_3$ **3** (positive mode).

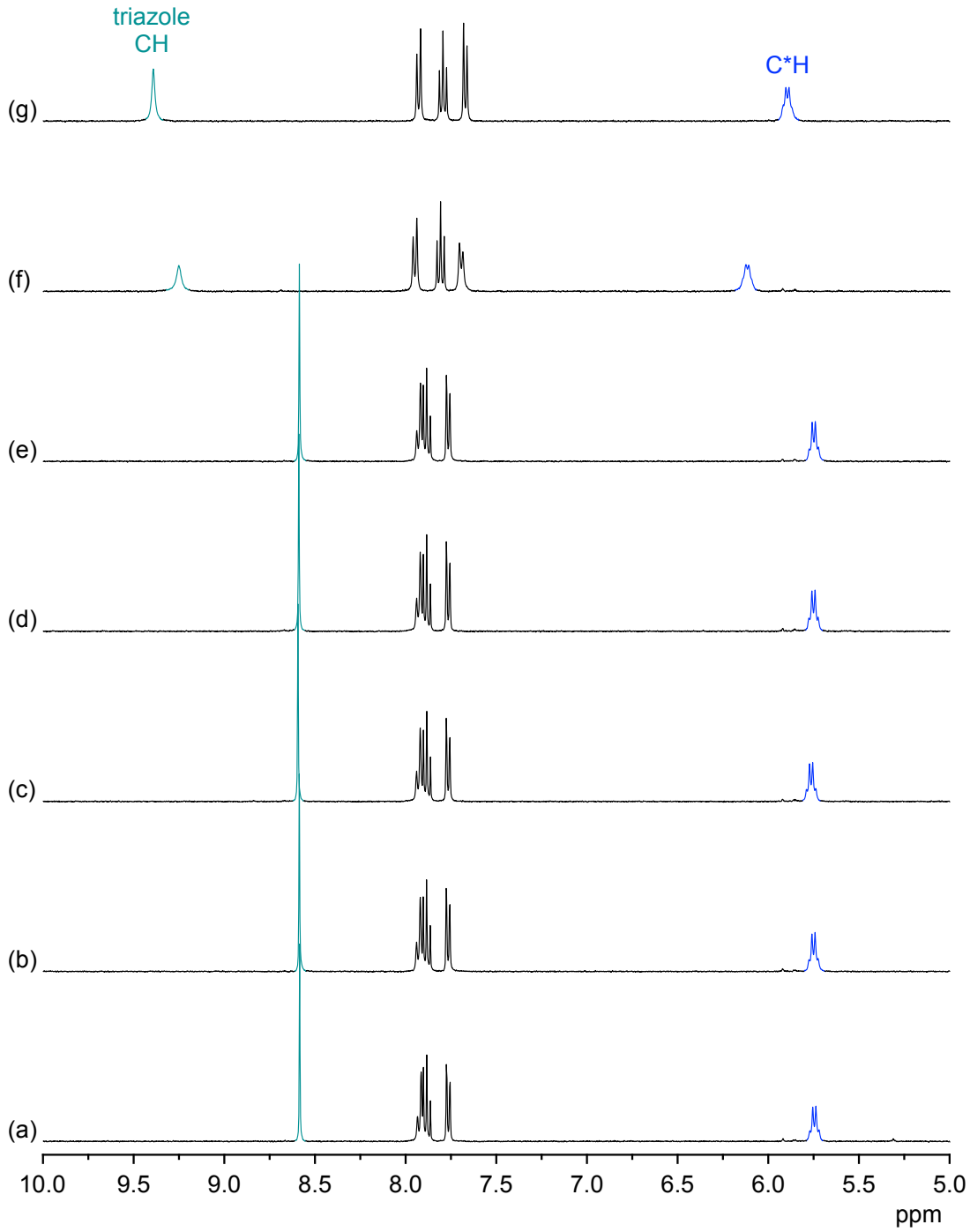


		m/z calcd.	*m/z exp.*
$[M+H]^+$	$C_{30}H_{27}N_{15}O_3 + H^+$	646.3	646.5
$[M+Na]^+$	$C_{30}H_{27}N_{15}O_3 + Na^+$	668.2	668.5
$[M+K]^+$	$C_{30}H_{27}N_{15}O_3 + K^+$	684.2	684.6

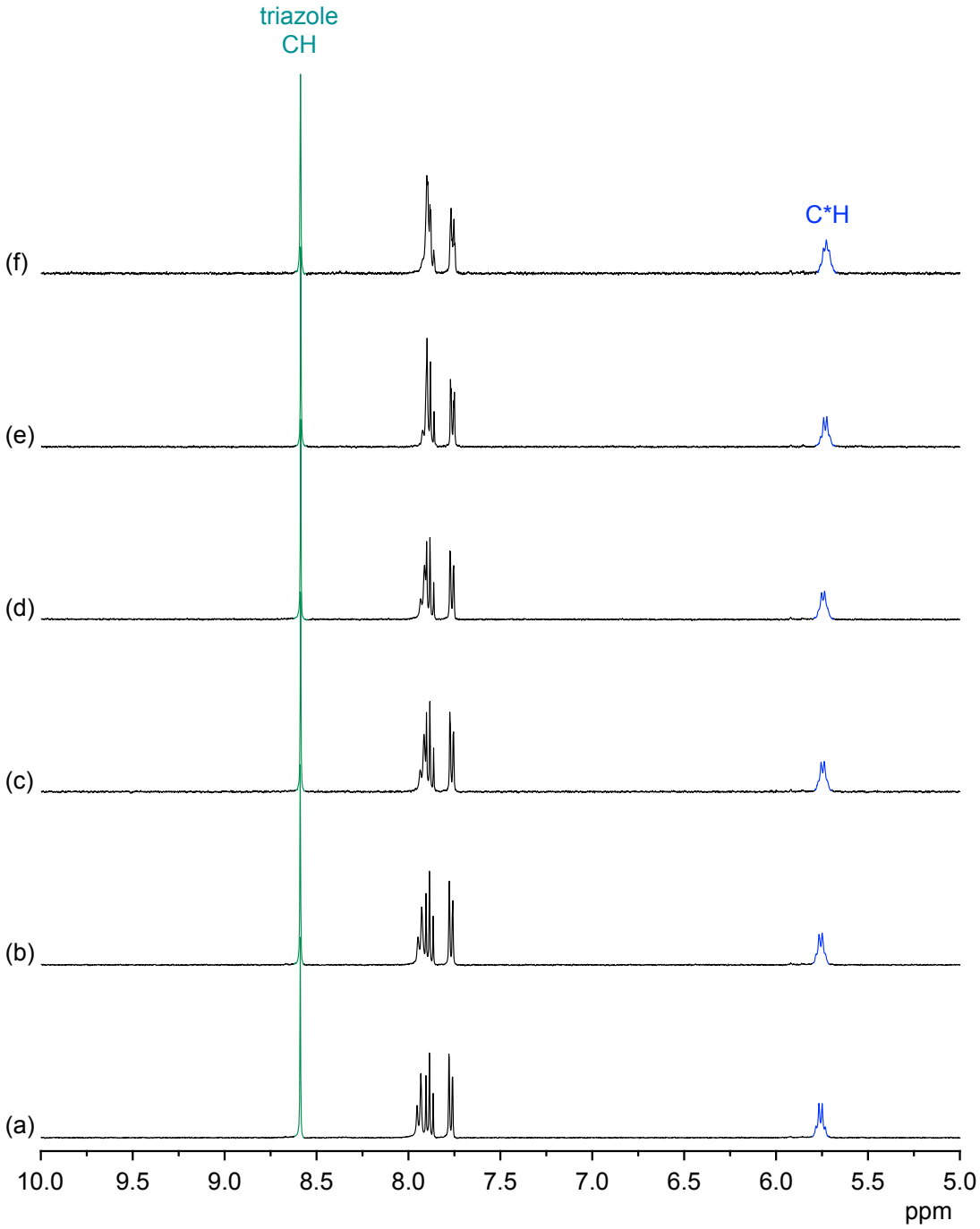
NOESY NMR Spectrum: **3** (1 mM) in DMSO-d_6 (mixing time 400 ms) (400 MHz).



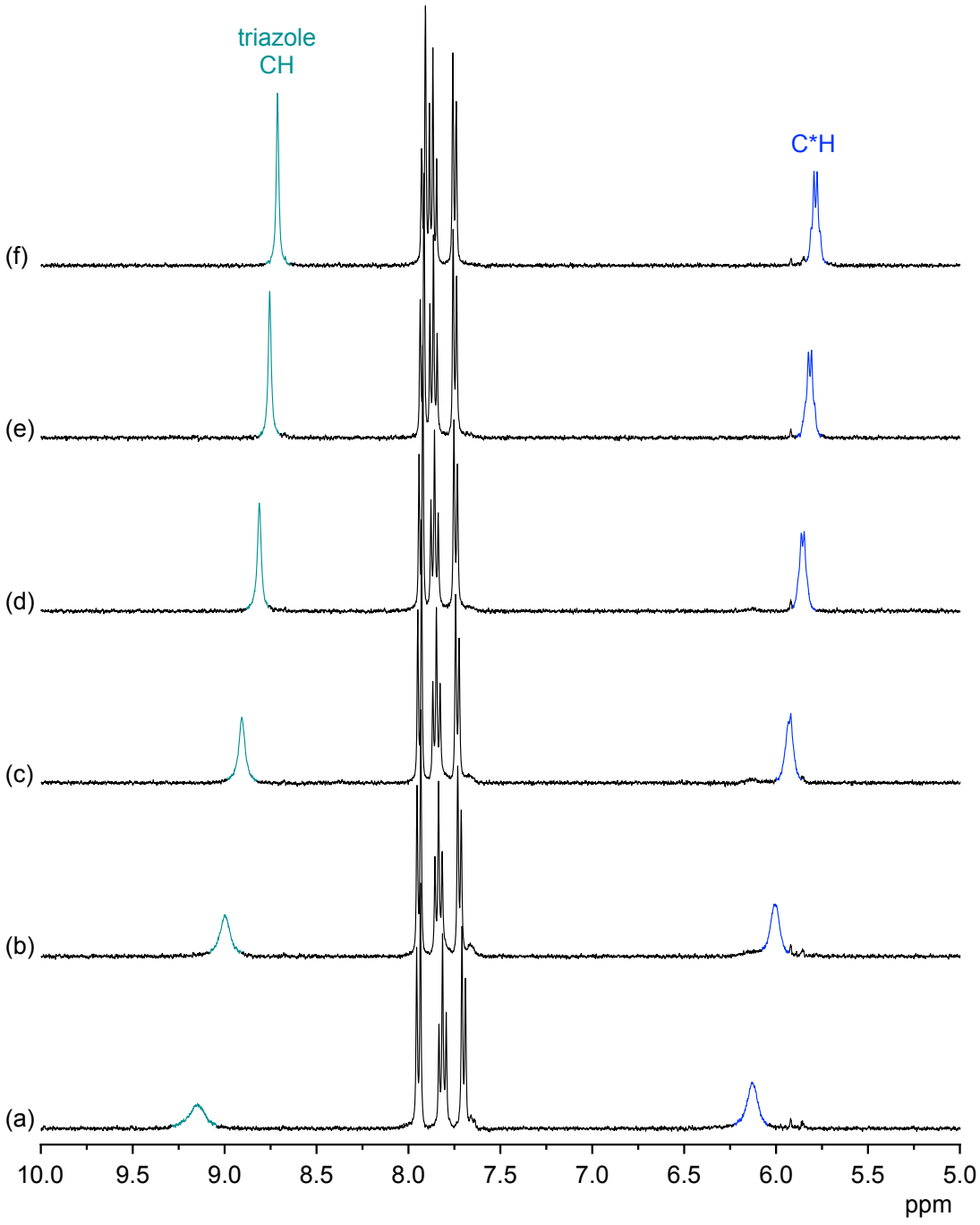
^{1}H NMR Spectra: **3** (1 mM) in 2.5 vol% D_2O/DMSO-d_6 in the absence (a) and the presence of 5 equiv of tetrabutylammonium iodide (b), bromide (c), chloride (d), nitrate (e), dihydrogenphosphate (f), and sulfate (g). The signals of the triazole CH, and C*H protons are marked in green and blue, respectively.



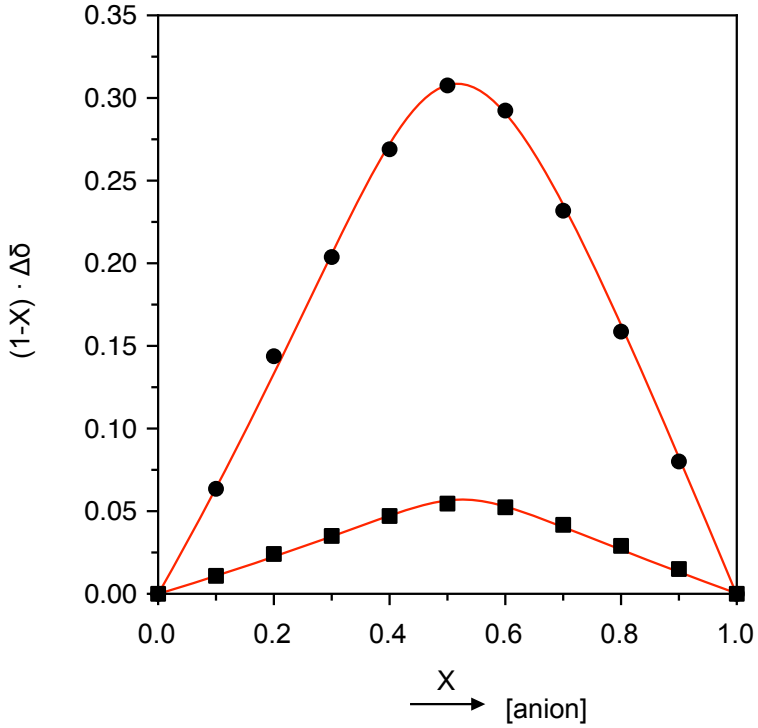
^{1}H NMR Spectra: **3** (1 mM) in D_2O/DMSO-d_6 mixtures with the D_2O content amounting to 0.03 vol% (a), 1 vol% (b), 2 vol% (c), 3 vol% (d) 4 vol% (e), and 5 vol% (f). The signals of the triazole CH, and C*H protons are marked in green and blue, respectively.



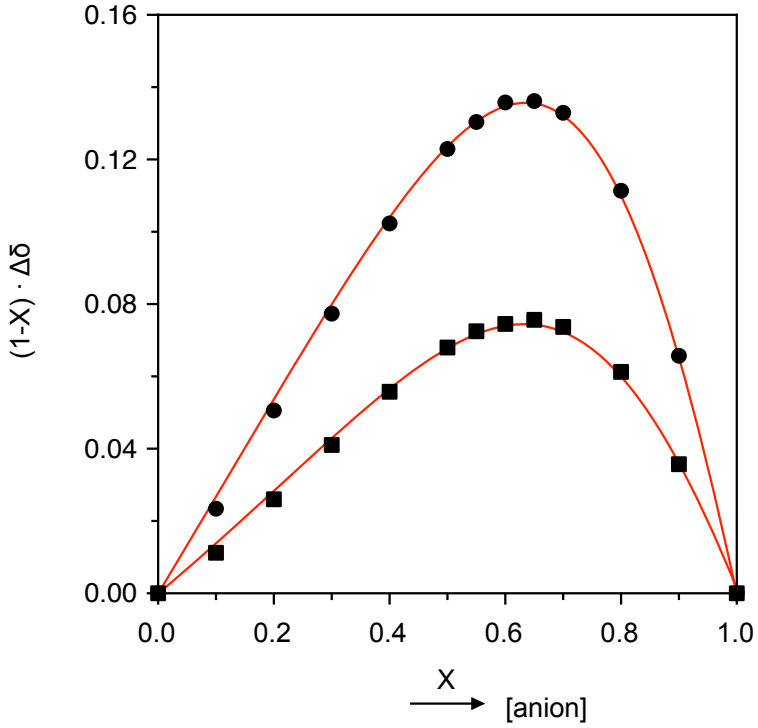
^{1}H NMR Spectra: **3** (1 mM) in the presence of 5 equiv of tetrabutylammonium dihydrogenphosphate in D_2O/DMSO-d_6 mixtures with the D_2O content amounting to 0.03 vol% (a), 1 vol% (b), 2 vol% (c), 3 vol% (d) 4 vol% (e), and 5 vol% (f). The signals of the triazole CH, and C*H protons are marked in green and blue, respectively.



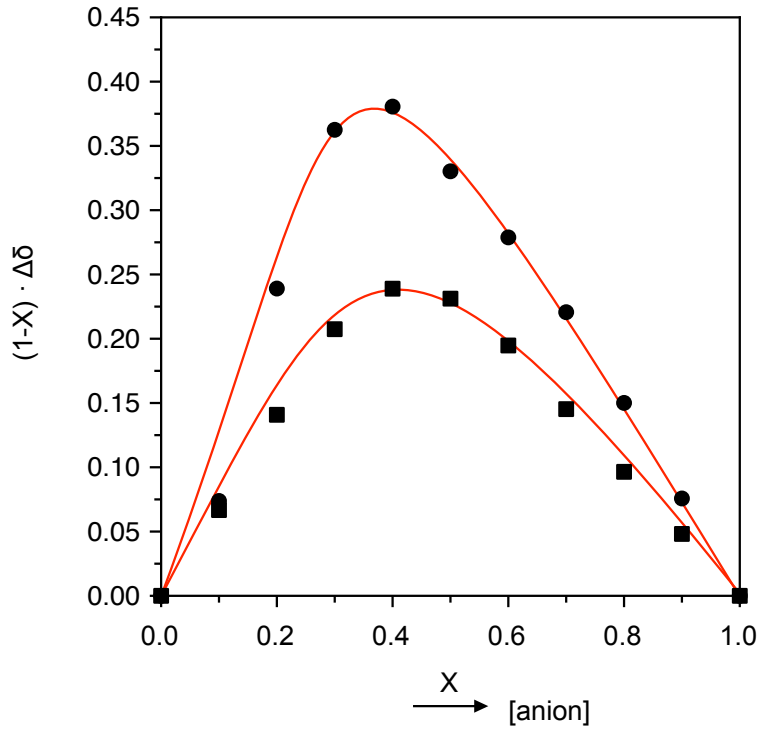
Job Plot: **3** + TBA sulfate (400 MHz, 2.5 vol% D_2O/DMSO-d_6). The circles indicate the shift of the triazole CH signal and the squares the one of the C*H signal.



Job Plot: **3** + TBA dihydrogenphosphate (400 MHz, 2.5 vol% D_2O/DMSO-d_6). The circles indicate the shift of the triazole CH signal and the squares the one of the C*H signal.

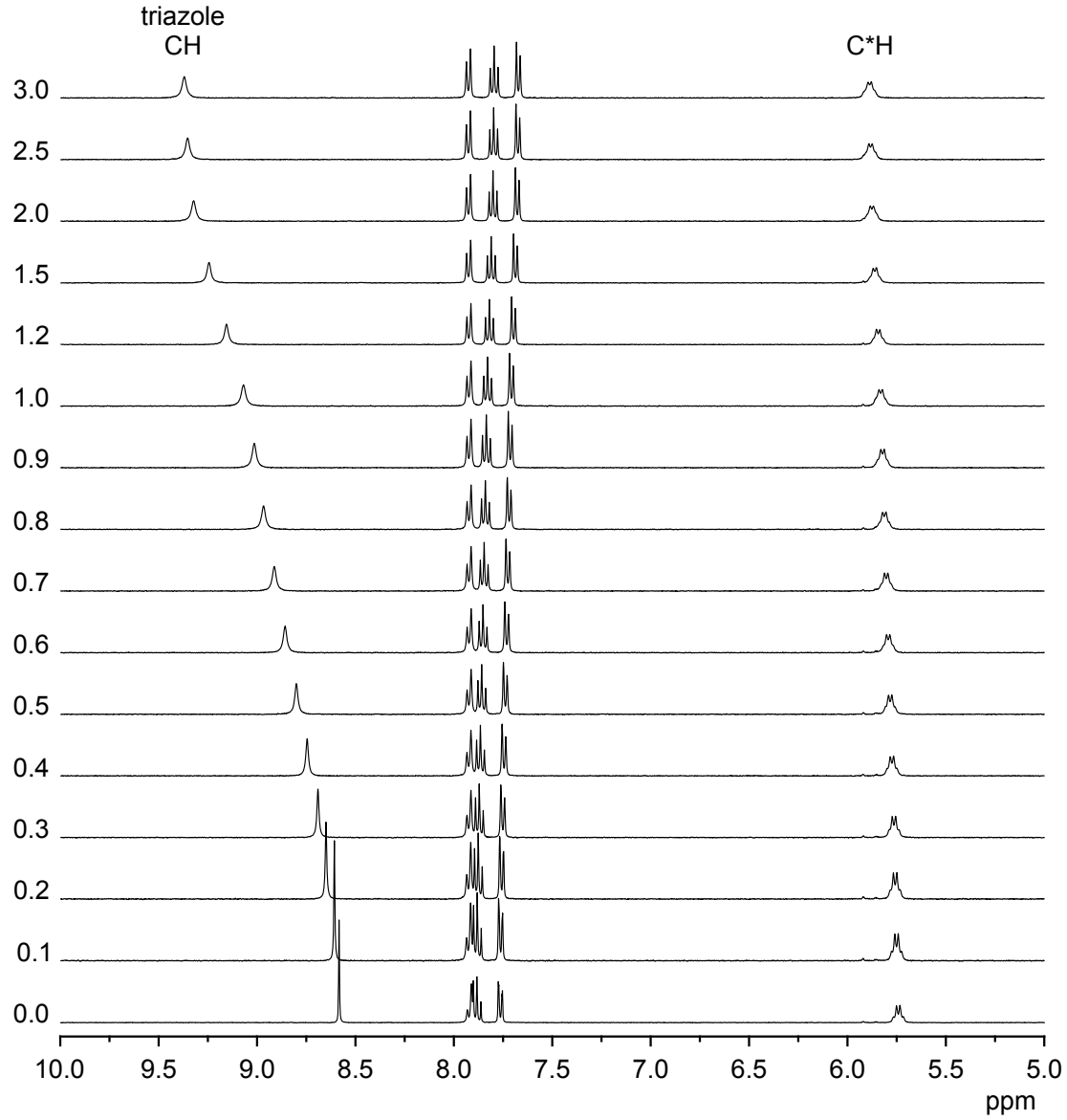


Job Plot: **3** + TBA trimetaphosphate (400 MHz, 2.5 vol% D_2O/DMSO-d_6). The circles indicate the shift of the triazole CH signal and the squares the one of the C*H signal.



NMR Titration: **3** + TBA sulfate.

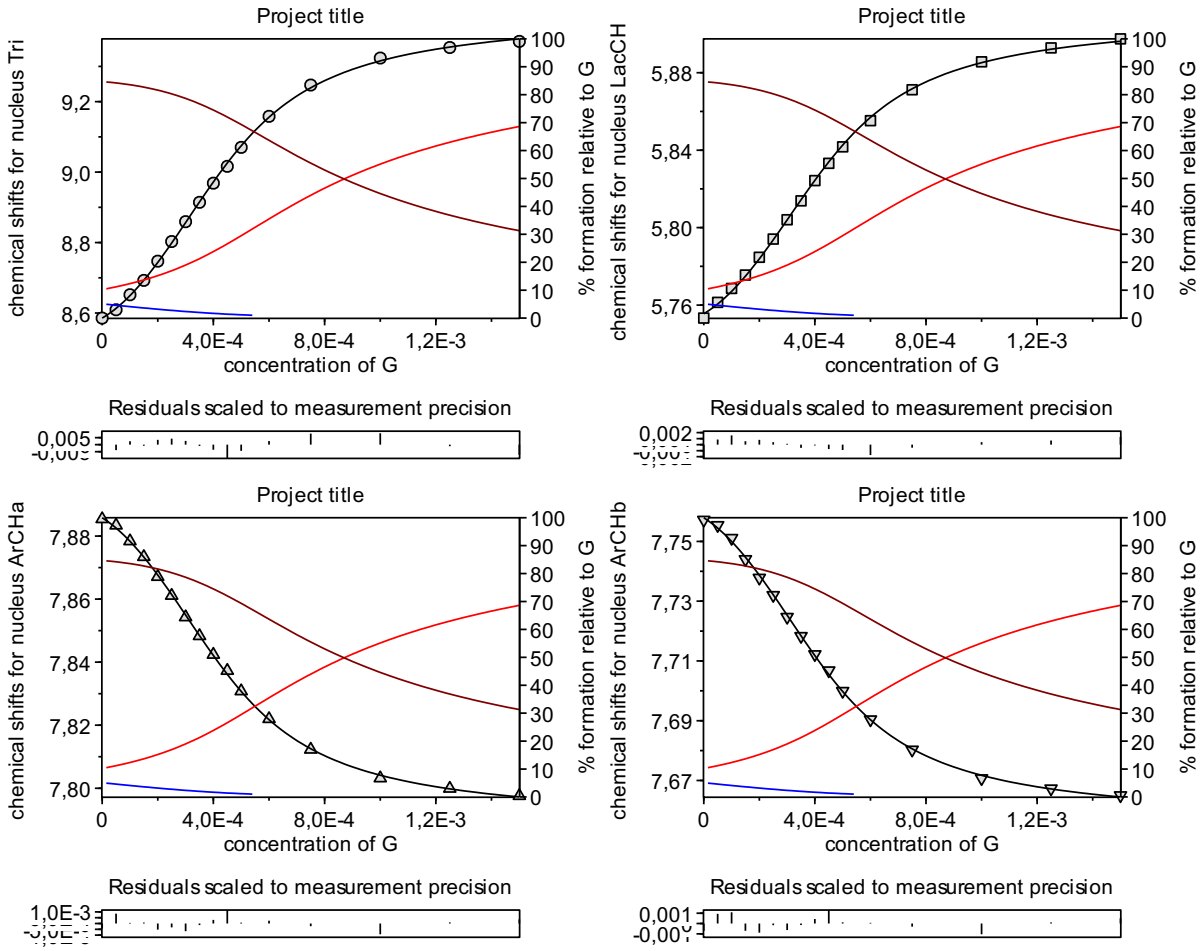
^{1}H NMR spectra of **3** (0.5 mM) in 2.5 vol% D_2O/DMSO-d_6 (400 MHz) containing the increasing equivalents of TBA sulfate specified to the left of each spectrum.



Binding isotherms and HypNMR outputs.

```
Converged in 1 iterations with sigma = 0.002798

                          standard
                 value    deviation   Comments
   log beta(GH2) 6.3      fixed
   log beta(GH)  4.2198   0.0124      4.22(1)
```



Evaluation of the stability constants of the sulfate complex with HypNMR was only possible by estimating and fixing the log β value representing the overall stability of the complex. To assess the effect of varying this parameter on the outcome of the regression, fitting was performed by using different log β values and the results were compared. The following table shows the dependence of log K_{11} and of the parameter sigma, describing the goodness of the fit, on the starting value of log β.

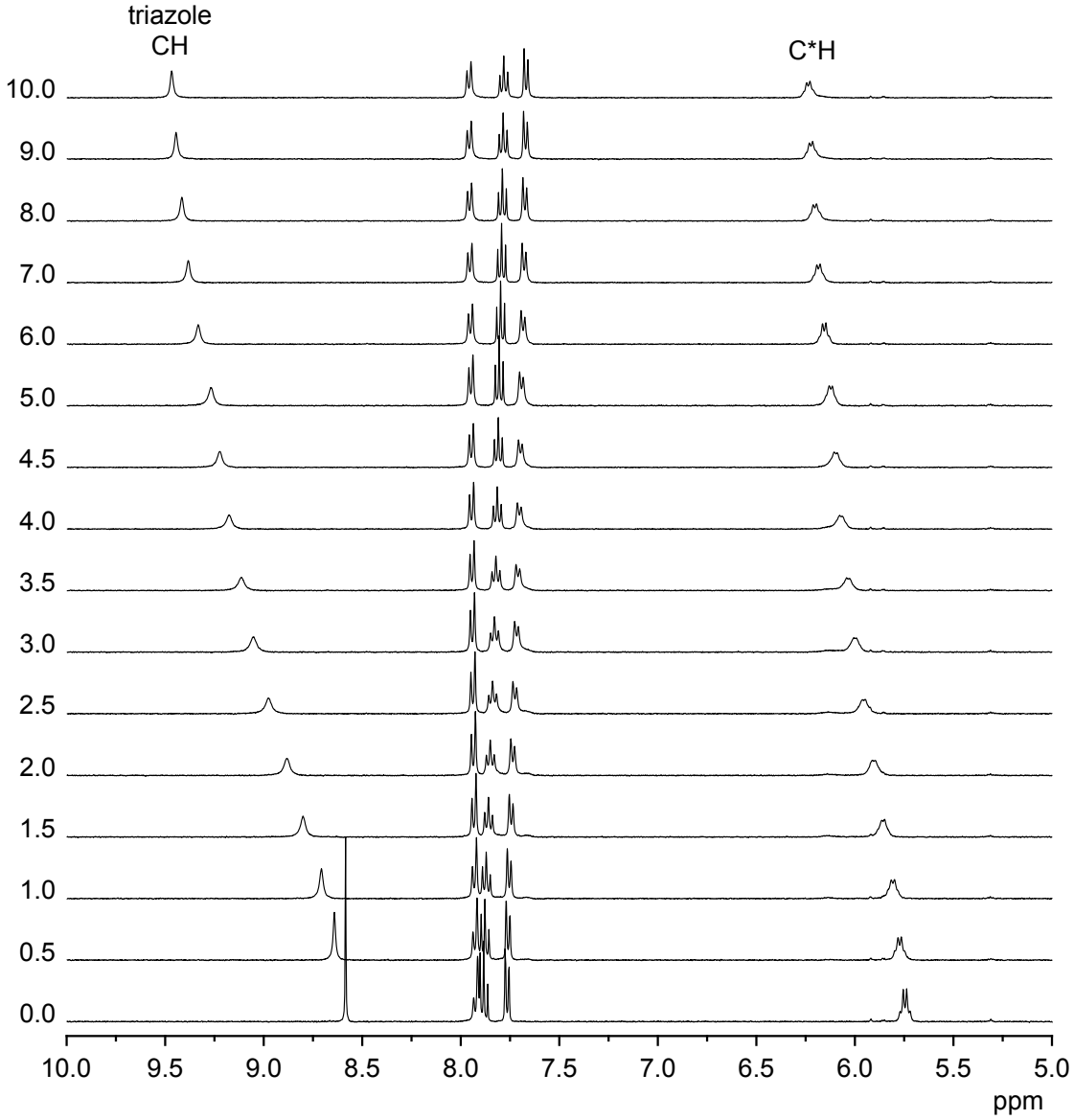
log β	log K_{11}	sigma
5.1	4.21	0.002672
5.3	4.21	0.002677
5.5	4.21	0.002685
5.7	4.21	0.002698
5.9	4.21	0.002718
6.1	4.21	0.002749
6.3	4.22	0.002798
7.0	4.26	0.003259
8.0	4.44	0.011096

No fitting was possible with log β < 5.1. The goodness of the fit becomes unacceptable with log β > 6.3. In the region 5.1 < log β < 6.3 the stability constant log K_{11} is practically unaffected by the initial choice of log β, rendering the estimation of this constant reliable. In the same region the sigma value only exhibits small variations so that the exact quantification of log K_{21} on the basis of this parameter is not possible. It is therefore safe to state than log K_{21} ranges between 0.9 and 2.1.

Note that also the signal of the methyl groups of **3** exhibits a minor shift in the ^{1}H NMR spectra during this titration. The resulting binding isotherm could, however, not be fitted to a reasonable binding model. As the methyl groups point away from the anion binding site, we attribute the shifts in their resonance to changes in solvation during the titration and we therefore neglected the corresponding binding isotherm in the data treatment.

NMR Titration: **3** + TBA dihydrogenphosphate.

^{1}H NMR spectra of **3** (0.5 mM) in 2.5 vol% D_2O/DMSO-d_6 (400 MHz) containing the increasing equivalents of TBA dihydrogenphosphate specified to the left of each spectrum.

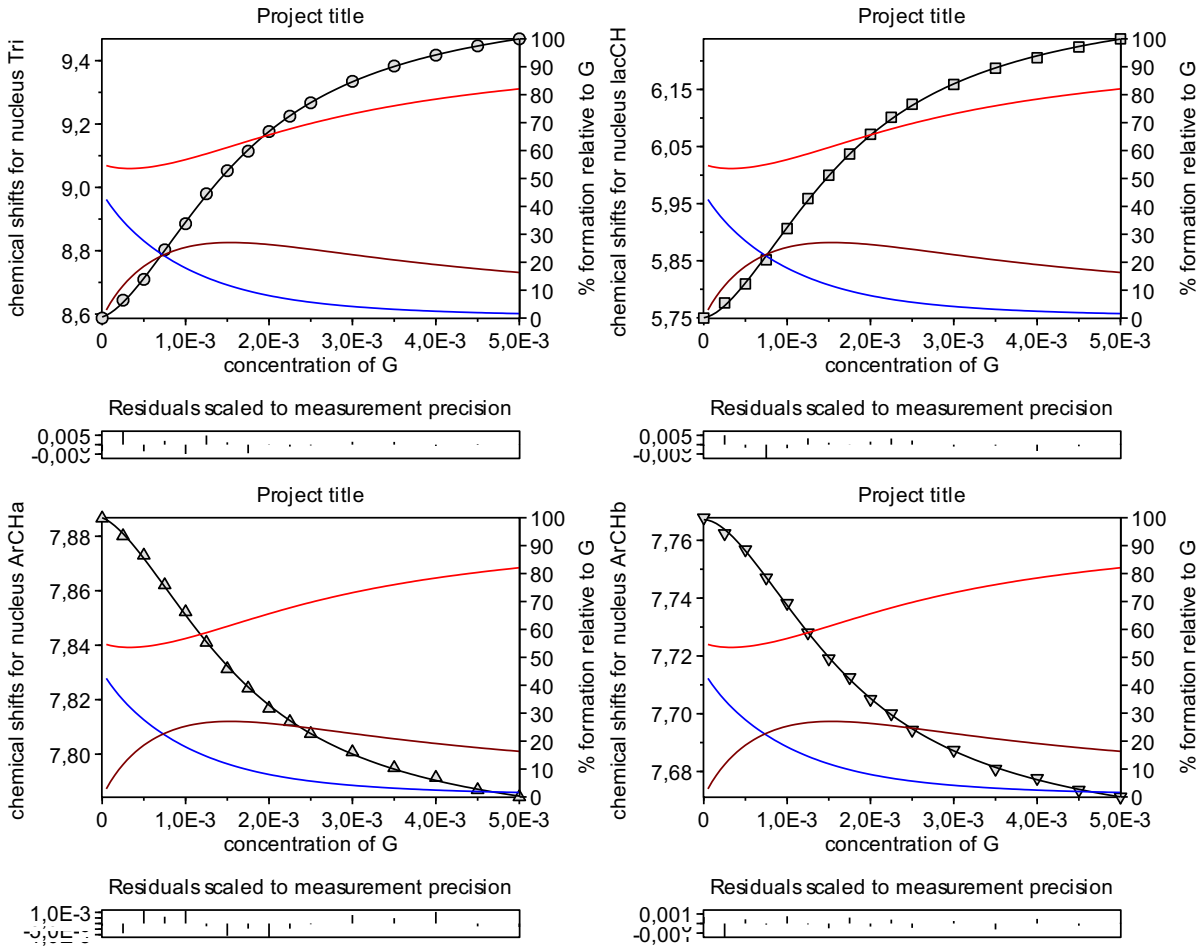


Binding isotherms and HypNMR outputs.

```
Converged in 1 iterations with sigma = 0.002448

                                standard
                    value       deviation    Comments
 1 log beta(GH)     3.2114      0.0997       3.21(1)
 2 log beta(G2H)    6.3066      0.0634       6.31(6)

Correlation coefficients between stability constants is 0.9858
```



In this case, both stability constants could be reliably fitted by using HypNMR without any initial assumptions. Only, the change of the resonance of the methyl signal of **3** was again neglected as in the sulfate titration.

Selected ITC Titrations:

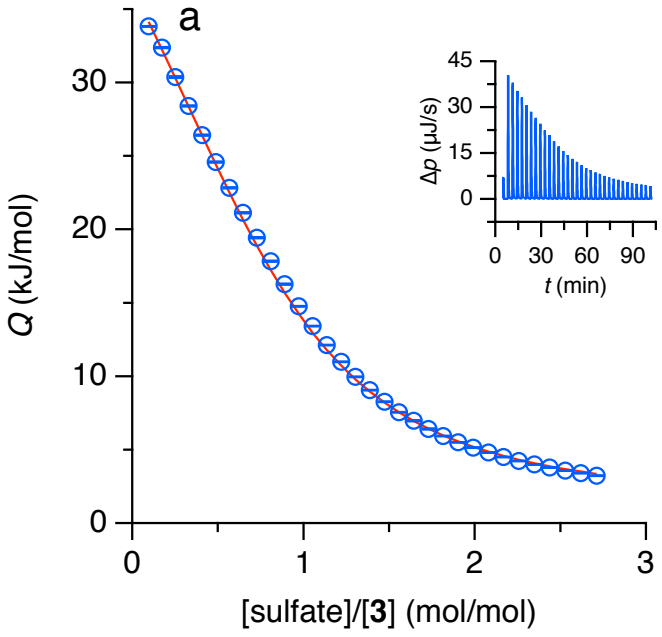
Table S1. Concentrations and experimental parameters of the individual titrations.

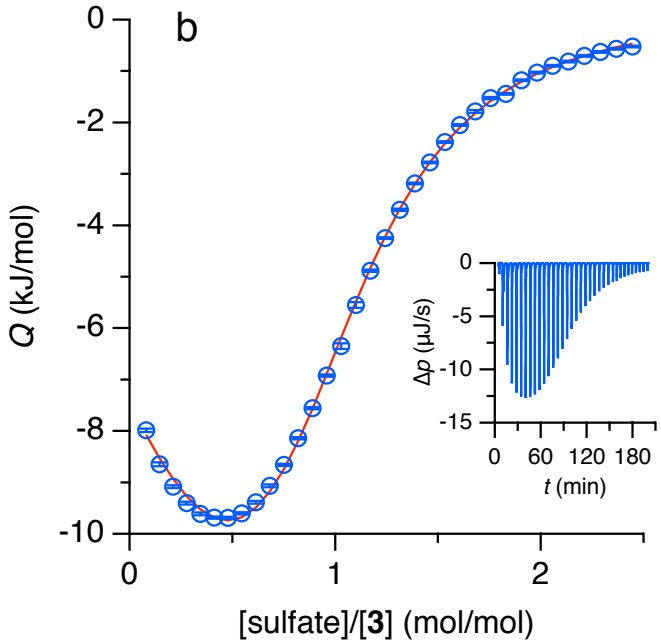
guest anion as TBA salt	vol% H_2O in H_2O/DMSO	*c*(**3**) / mM	*c*(salt) / mM	injection volume / µL	spacing time / s
	0.03	0.25	3.6	8	180
sulfate	2.5	0.37	4.4	8	360
	5	0.25	3.8	8	360
	0.03	0.51	19.0	8[a]	200
DHP	2.5	0.47	33.7	8[a]	180
	5	0.52	38.5	8	180
HPP	2.5	0.27	4.9	8	200
TMP	2.5	0.51	5.1	8	360

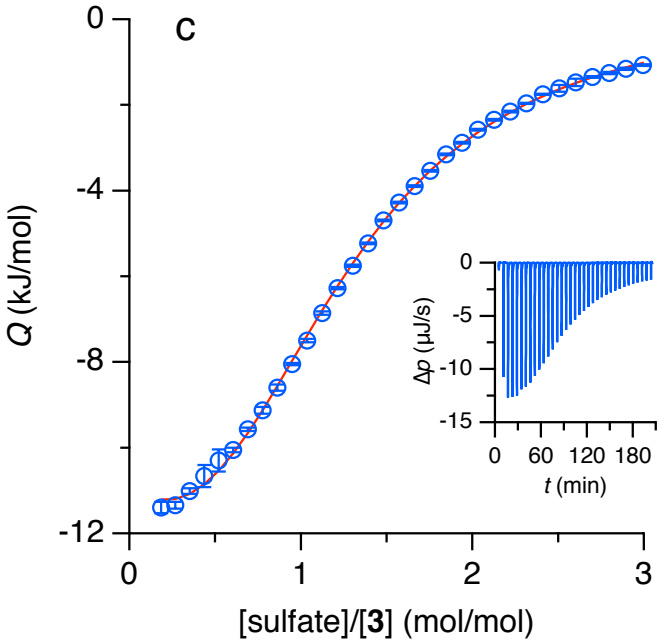
[a] For the first 4 injections an injection volume of 4 µL was used.

Thermograms were baseline-corrected with NITPIC,[1] and the resulting binding isotherms were fitted with SEDPHAT.[2-4] Circles in isotherms denote experimental data, and lines represent fits based on the binding models explained in the main text.

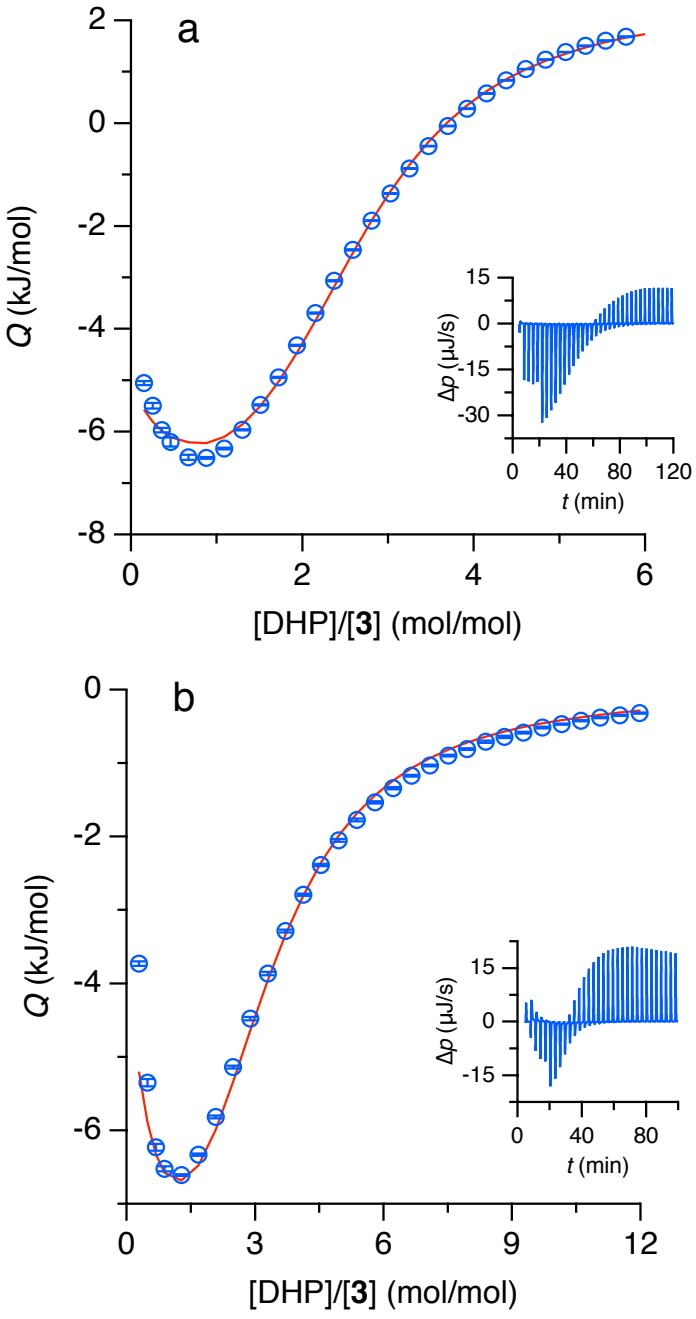
Titration of **3** with TBA sulfate (a: 0.03 vol% H_2O in DMSO, b: 2.5 vol% H_2O in DMSO, c: 5 vol% H_2O in DMSO).

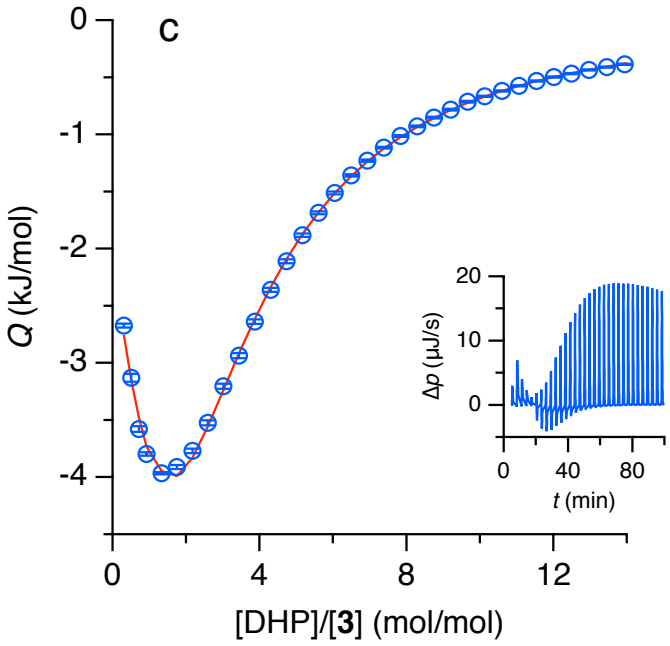



b
0
-2
-4
-6
-8
-10
Q (kJ/mol)
0
1
2
[sulfate]/[3] (mol/mol)
0
-5
-10
-15
Δp (μJ/s)
0
60
120
180
t (min)

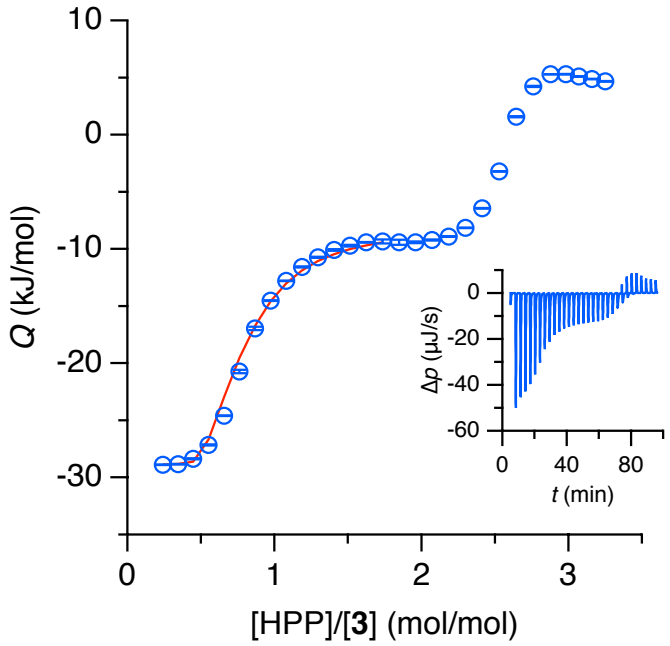

c
0
-4
-8
-12
Q (kJ/mol)
0
1
2
3
[sulfate]/[3] (mol/mol)
0
-5
-10
-15
Δp (μJ/s)
0
60
120
180
t (min)

Titration of **3** with TBA dihydrogenphosphate (DHP) (a: 0.03 vol% H_2O in DMSO, b: 2.5 vol% H_2O in DMSO, c: 5 vol% H_2O in DMSO).

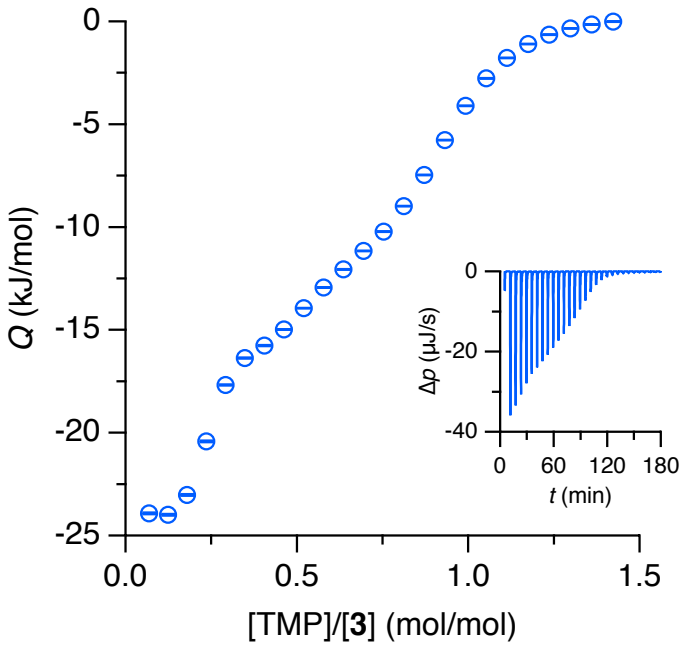




Titration of **3** with TBA hydrogenpyrophosphate (HPP) (2.5 vol% H_2O in DMSO).



Titration of **3** with TBA trimetaphosphate (TMP) (2.5 vol% H_2O in DMSO).



Crystal Structures

Structure of 3·5DMSO·0.35H_2O: **3**·5DMSO·0.35H_2O was crystallized as colourless plates by slow evaporation of an acetone/DMSO solution of **3**. The single crystal X-ray data was collected at 150.0(1) K on a Rigaku/Oxford diffraction Xcalibur/Gemini dual wavelength diffractometer with a Cu-Kα (λ = 1.54184 Å) radiation X-ray source. Program *CrysAlisPro*[5] was used for the data collection and reduction. The intensities were corrected for absorption using analytical face index absorption correction method[6] for all the data.

Both structures were solved with direct methods (*SHELXS-2014/7*[8]) and refined by full-matrix least squares on F^2 using *SHELXL-2014/7*[8] program. Anisotropic displacement parameters were assigned to non-H atoms. All the hydrogen atoms were refined using riding models. The hydrogen atoms from O8 were not localized.

Crystal data: 0.58×0.35×0.03 mm, $C_{40}H_{57}N_{15}O_{8.35}S_5$, M = 1041.91, orthorhombic, space group $P2_12_12_1$, a = 9.3172(2) Å, b = 18.6791(4) Å, c = 29.1423(5) Å, V = 5071.84(18) Å^3, Z = 4, μ(Mo-Kα) = 2.649 mm^{-1}, D_{calc} = 1.362 g/cm^3, F(000) = 2192, 15405 reflections measured (3.847° ≤ Θ ≤ 62.663°), 7663 unique (R_{int} = 0.0311, R_{sigma} = 0.0403) which were used in all calculations. Final R indices ($I > 2\sigma(I)$): R_1= 0.0327, wR_2 = 0.0821, R indices (all data): R_1= 0.0350, wR_2 = 0.0843. *GOF* = 1.033 for 668 parameters and 18 restraints, largest diff. peak and hole 0.365/–0.293 eÅ^{-3} with Flack parameter 0.005(8). CCDC-1505706 contains the supplementary data for this structure.

Structure of 3·1.5TBADHP·2.5DMSO·1.7H_2O: Colourless prisms of **3**·1.5TBADHP·2.5DMSO·1.7H_2O were obtained by slow evaporation of acetone/DMSO solution of **3** and TBADHP. The single crystal X-ray data was collected at 120.0(1) K on a Rigaku/Agilent Super-Nova dual wavelength diffractometer with a micro-focus X-ray source and multilayer optics monochromatised Cu-Kα (λ = 1.54184 Å) radiation. For data collection, reduction, and absorption correction, see above.

The solution of the structure varied by using *SIR2014*[7] for the direct methods and the handling of one hydrogen atom. The one hydrogen (H5O) between dihydrogen phosphates was located from the difference-Fourier map and refined with U_{eq}(H) of 1.5 U_{eq}(O) and with O-H distance restrained to value of 1.00 Å (s = 0.01). The compound is pure enantiomer and crystallized in a chiral space group,

but due to weak disordered data the absolute structure could not be determined reliably.

Crystal data: 0.30×0.23×0.15 mm, $C_{118}H_{198}N_{33}O_{26.40}P_3S_5$, M = 2754.69, orthorhombic, space group $P2_12_12$, a = 32.0131(5) Å, b = 15.5735(2) Å, c = 14.7710(4) Å, V = 7634.2(2) Å^3, Z = 2, μ(Cu-Kα) = 1.656 mm^{-1}, D_{calc} = 1.242 g/cm^3, F(000) = 2946, 27356 reflections measured (3.960° ≤ Θ ≤ 68.246°), 13466 unique (R_{int} = 0.0300, R_{sigma} = 0.0394) which were used in all calculations. Final R indices ($I > 2\sigma(I)$): R_1= 0.1279, wR_2 = 0.3742, R indices (all data): R_1= 0.1365, wR_2 = 0.3923. GOF = 1.760 for 965 parameters and 511 restraints, largest diff. peak and hole 1.15/–0.64 eÅ^{-3} with Flack parameter 0.037(8). CCDC-1504361 contains the supplementary data for this structure.

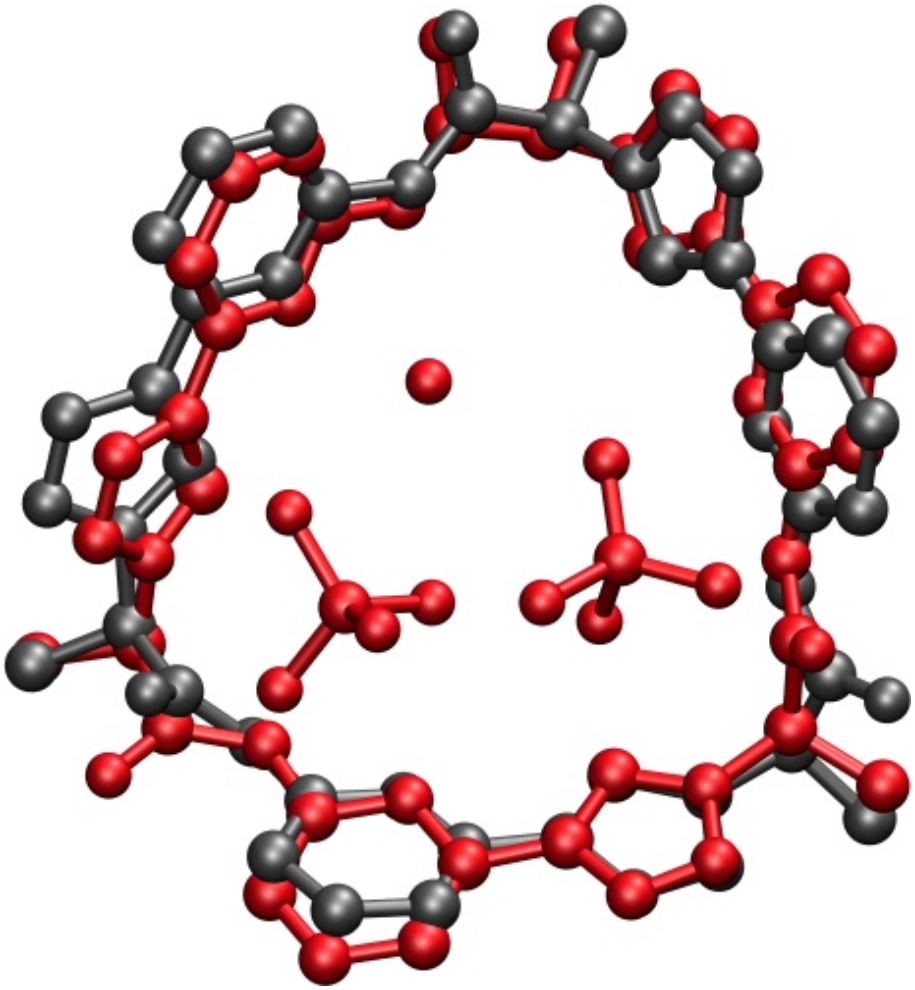


Figure S1: Overlay of the molecular structures of pseudopeptide **3** in the crystals **3**·5DMSO·0.35H_2O (black) and **3**·1.5TBADHP·2.5DMSO·1.7H_2O (red).

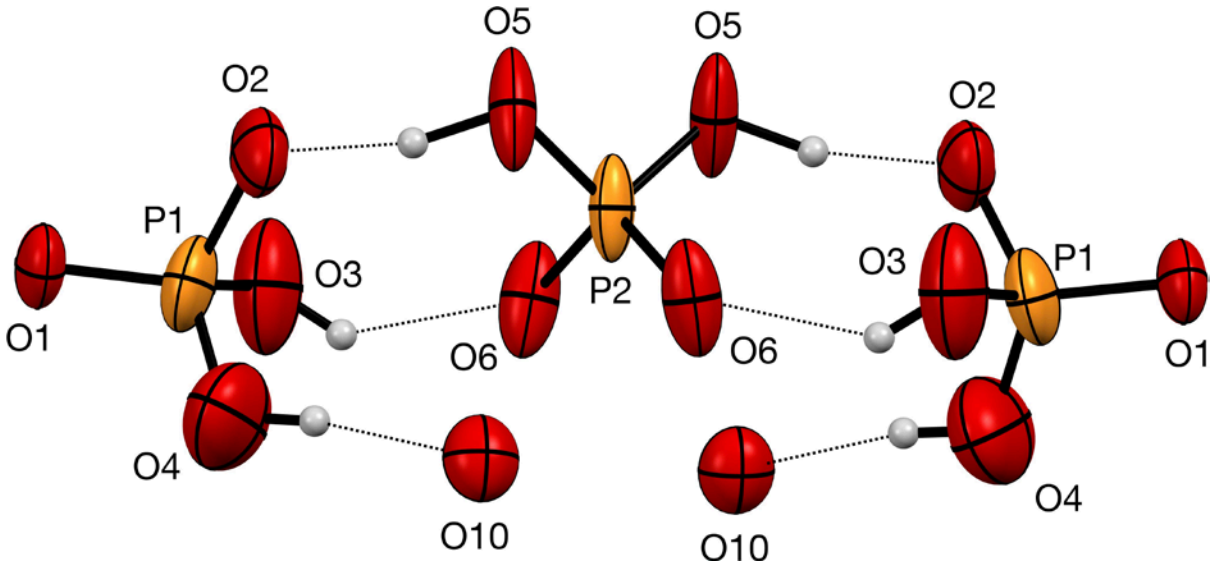


Figure S2: Detailed representation of the hydrogen bonding pattern in the DHP trimer of **3**·1.5TBADHP·2.5DMSO·1.7H_2O.

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