Carboxylate-Based Receptors for the Recognition of Carbohydrates in Organic and Aqueous Media

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1. Syntheses of the compounds **9**, **10**, **11a** and **11b**.

^{1}H and ^{13}C NMR spectra were measured using a 400 and 500 MHz spectrometer; chemical shifts are reported in ppm downfield to TMS as internal standard. Analytical TLC was carried out on silica gel 60 F_{254} plates employing a methanol-chloroform 1:7 (v/v) as the mobile phase. Melting points are uncorrected.

1-[Bis(*N*,*N*-diethoxycarbonylmethyl)aminomethyl]-3,5-bis[(6-methyl-pyridin-2-yl)-aminomethyl]-2,4,6-trimethyl-benzene (9): A mixture of 1,3,5-tris(bromomethyl)-2,4,6-trimethyl-benzene[12] (3.00 g, 7.50 mmol), K_2CO_3 (3.15 g, 22.50 mmol) and iminodiacetic acid diethyl ester (1.41 g, 7.50 mmol) in CH_3CN (120 mL) was stirred at room temperature for 45 min. Then 2-amino-6-methyl-pyridine (1.62 g, 15 mmol) in THF (50 mL) were added, and the mixture was stirred at room temperature for 48 h. After filtration and evaporation of solvents, the crude product was purified by column chromatography (ethyl acetate/toluene 1:1 v/v). Yield 33%. M.p. 116-118 °C. ^{1}H-NMR (400 MHz, $CDCl_3$): δ = 7.36 (t, 2H, *J*= 7.8 Hz), 6.47 (d, 2H, *J* = 7.1 Hz), 6.26 (d, 2H, *J* = 8.3 Hz), 4.38 (d, 4H, *J* = 4.3 Hz), 4.23 (t, 2H, *J* = 4.3 Hz), 4.12 (q, 4H, *J* = 7.1 Hz), 4.04 (s, 2H), 3.51 (s, 4H), 2.44 (s, 6H), 2.38 (s, 9H). 1.25 (t, 6H, *J* = 7.1 Hz). ^{13}C-NMR (100 MHz, $CDCl_3$): δ = 171.4, 158.2, 157.1, 138.0, 137.8, 136.6, 133.4, 132.8, 112.2, 102.9, 60.3, 53.1, 52.3, 41.8, 24.4, 15.9, 14.2. HR-MS calcd for $C_{32}H_{43}N_5O_4$ 561.3313; found: 561.3315. $R_{f=}$ 0.74 (methanol-chloroform 1:7 v/v).

Diacid 10: Diester **9** (103 mg, 0.18 mmol) was dissolved in a solution containing THF (10 mL), MeOH (10 mL), H_2O (10 mL) and NaOH (16 mg, 0.40 mmol). The solution was stirred at room temperature for 15 h. The organic solvents were removed under reduced pressure and the water phase was acidified with 5% HCl. The solution was evaporated to dryness and the residue was suspended in EtOH. After filtration and evaporation of EtOH, the obtained

powder was dried, and then suspended in $CHCl_3$ containing NEt_3. This mixture was stirred at room temperature for 45 min and then evaporated to dryness. The residue was washed with small amounts of water (separation of triethylamine hydrochloride) and dried to obtaine **9** as a white powder.

Yield 93%. M.p. 161-162 °C. ^{1}H-NMR (400 MHz, DMSO): δ = 7.24 (t, 2H, *J* = 7.8 Hz), 6.36 (d, 2H, *J* = 7.1 Hz), 6.23 (d, 2H, *J* = 8.3 Hz), 4.39 (d, 4H, *J* = 4.3 Hz), 3.95 (s, 2H), 3.41 (s, 4H), 2.44 (s, 6H), 2.34 (s, 3H), 2.30 (s, 6H). ^{13}C-NMR (100 MHz, DMSO): δ = 171.7, 158.1, 155.3, 137.1, 136.8, 136.3, 133.6, 132.1, 110.3, 105.5, 52.6, 51.7, 40.4, 24.1, 15.7. HR-MS calcd for $C_{28}H_{33}N_5O_3$ [M-H_2O] 487.2591; found: 487.2584. R_f = 0.20 (methanol-chloroform 1:7 v/v). **Sodium salt 11a**: HR-MS calcd for $C_{28}H_{34}N_5Na_2O_4$ $[M + H]^+$ 550.2406; found: 550.2403.

Bis(tetramethylammonium) salt 11b: Two equiv. of a 0.1 M solution of Me_4NOH in methanol/2-propanol were added to the diacid in methanol. The resulting solution was stirred, evaporated to dryness under reduced pressure, and the solid salt was dried in vacuo. HR-MS calcd for $C_{28}H_{34}N_5O_4$ $[M-(NMe_4)_2 + H]^-$ 504.2610; found: 504.2611.

Binding Studies. ^{1}H NMR titrations were performed at 298 K in $CDCl_3$ stored over activated molecular sieves and deacidified with Al_2O_3 (for each titration 10-20 samples were prepared). The titration data were analyzed by nonlinear regression analysis using the Hostest 5.6 program.[13] For each system at least 3 titrations were carried out. The used concentration ranges are given in the legend of Figures 2-7 and S1-S4.

2. ^{1}H NMR titration of **11b** with β-glucopyranoside **12b** in $CDCl_3$ (chemical shifts of the CH_3 and the pyridine CH resonances).

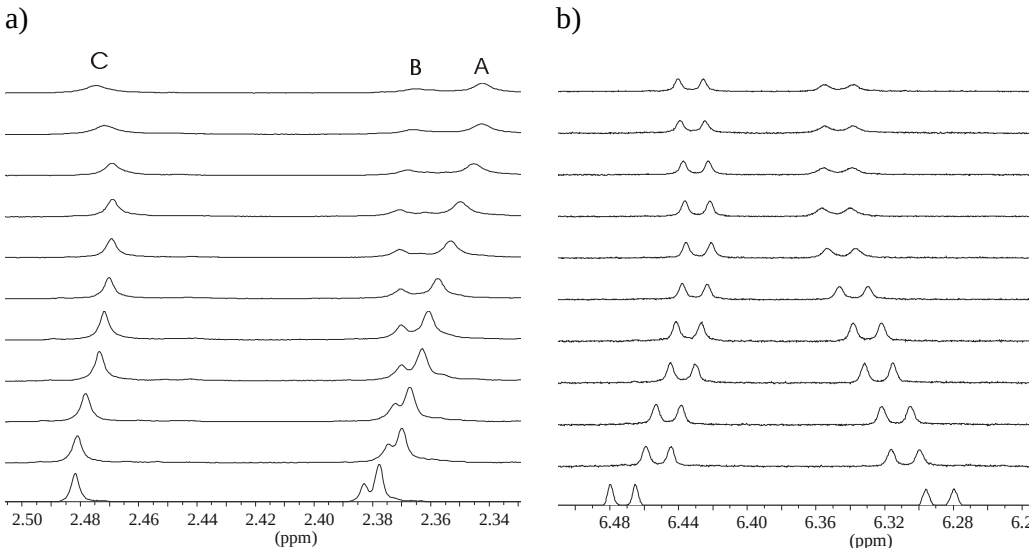


FIGURE S1. Partial ^{1}H NMR spectra (500 MHz, $CDCl_3$, 25 °C) of **11b** after addition of (from bottom to top) 0, 0.34, 0.51, 0.69, 0.86, 1.21, 1.55, 2.16, 3.03, 3.89 and 4.32 equiv of **12b** ([**11b**] = 0.85 mM). (a) Chemical shifts of the CH_3 resonances. (b) Chemical shifts of the pyridine CH resonances, HC-3 and HC-5.

3. ^{1}H NMR Titration of **11b** with α-glucopyranoside **13** in $CDCl_3$.

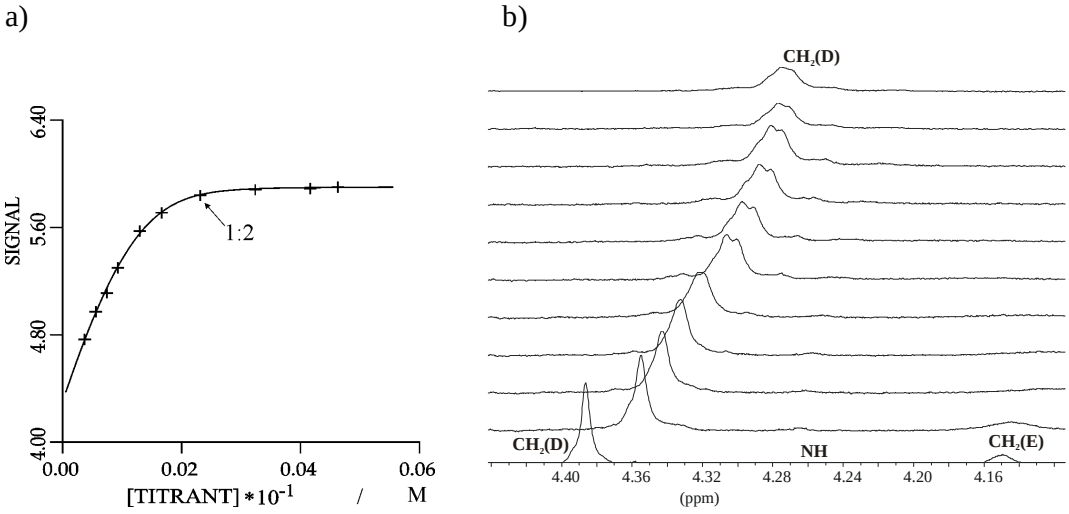


FIGURE S2. (a) Plot of the observed (**x**) and calculated (—) downfield chemical shifts of the NH resonances of **11b** as a function of added α-glucopyranoside **13**; [**11b**] = 0.85 mM; Equiv of **13** = 0.31, 0.47, 0.63, 0.79, 1.10, 1.42, 1.98, 2.77, 3.56, and 3.96. The [receptor]:[sugar] ratio is marked. (b) Partial ^{1}H NMR spectra (500 MHz, $CDCl_3$, 25 °C) of **11b** (CH_2 resonances, D and E, are shown) after addition of (from bottom to top) 0, 0.31, 0.47, 0.63, 0.79, 1.10, 1.42, 1.98, 2.77, 3.56, and 3.96. equiv of **13**.

4. ^{1}H NMR Titration of receptor **9** with α-glucopyranoside **13** in $CDCl_3$.

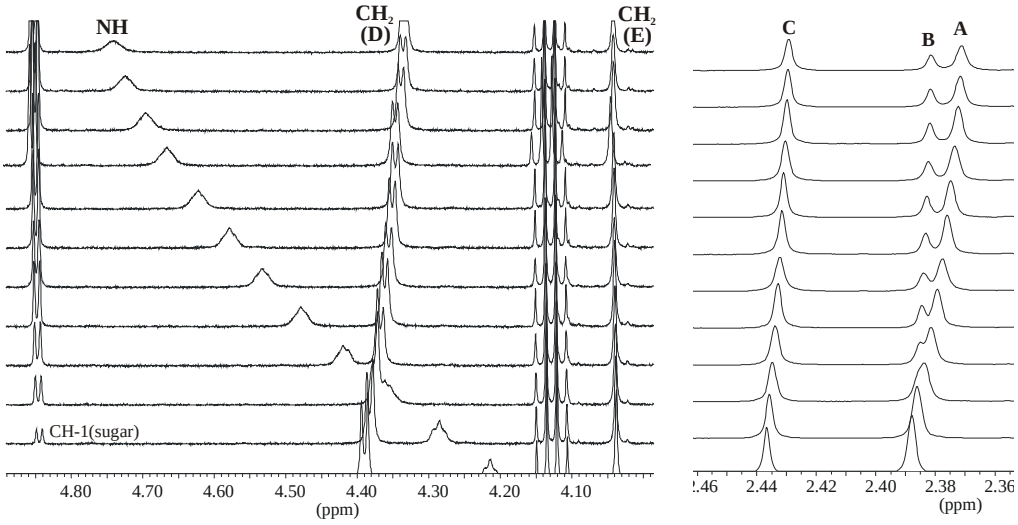


FIGURE S3. ^{1}H NMR titration of receptor **9** with α-glucopyranoside **13** in $CDCl_3$. Partial ^{1}H NMR spectra of **9** after addition of (from bottom to top) 0, 0.50, 1.01, 1.51, 2.02, 2.52, 3.03, 3.53, 4.04, 4.54, 5.05 and 6.00 equiv of **13** ([**9**] = 0.91 mM).

5. ^{1}H NMR titration of receptor **11a** with cellobiose **14** in H_2O/D_2O (chemical shifts of the CH_3 resonances)

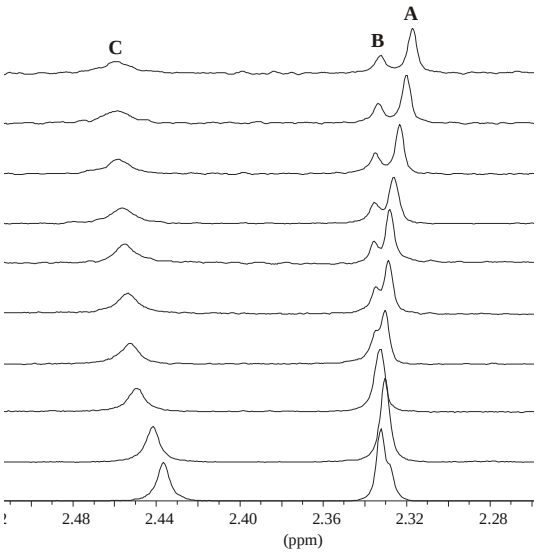


FIGURE S4.) Partial ^{1}H NMR spectra of **11a** in H_2O/D_2O (93:7 v/v) after addition of (from bottom to top) 0, 29, 44, 58, 87, 116, 145, 174, 233, 291 and 349 equiv of **14** ([**11a**] = 0.72 mM).

6. ^{1}H and ^{13}C NMR spectra of compounds **9**, **10**, **11a** and **11b**.

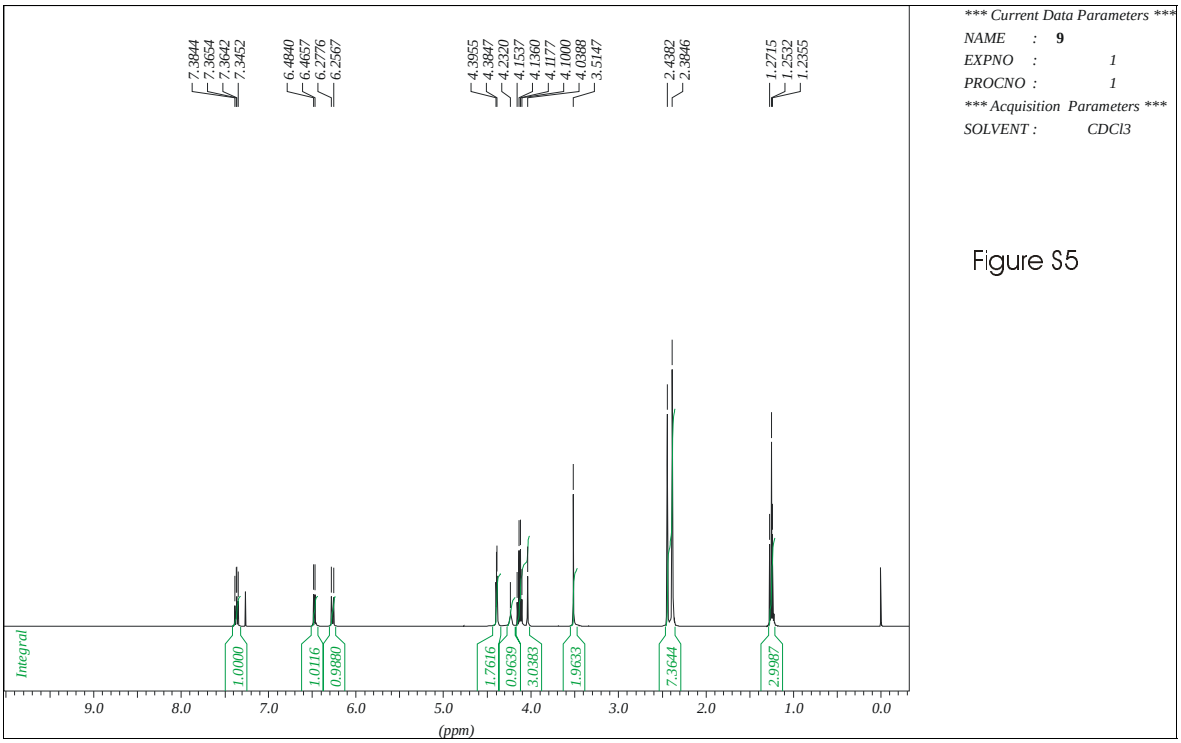
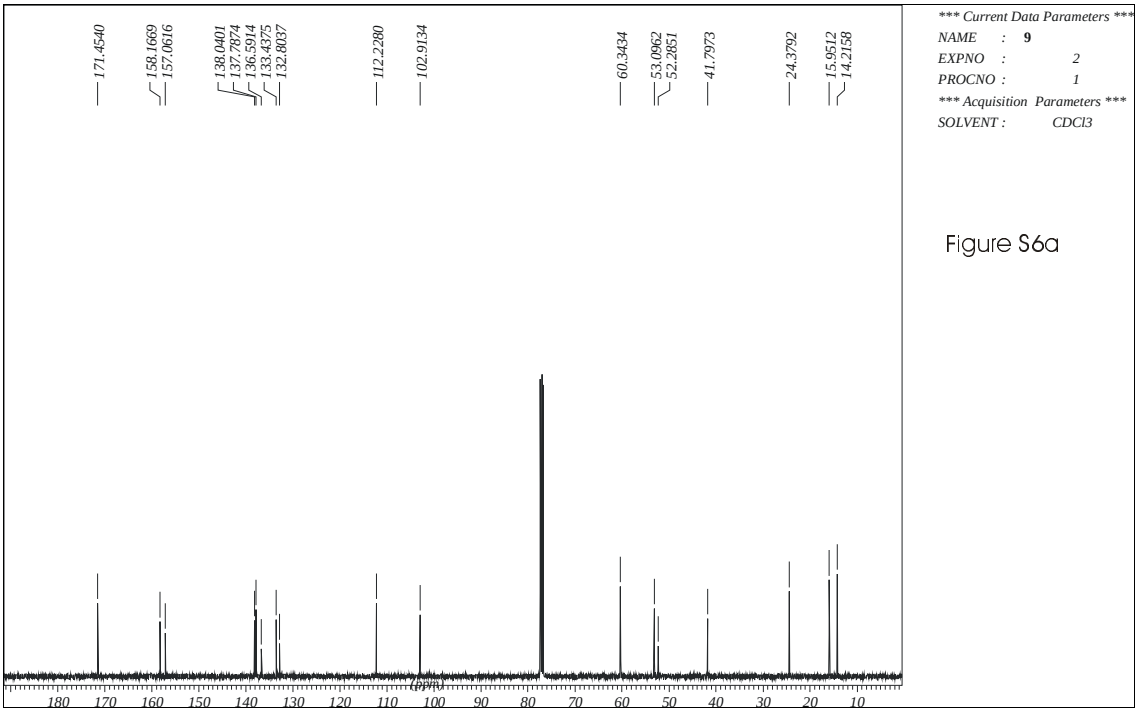


FIGURE S5. ^{1}H NMR spectrum of **9** in $CDCl_3$.



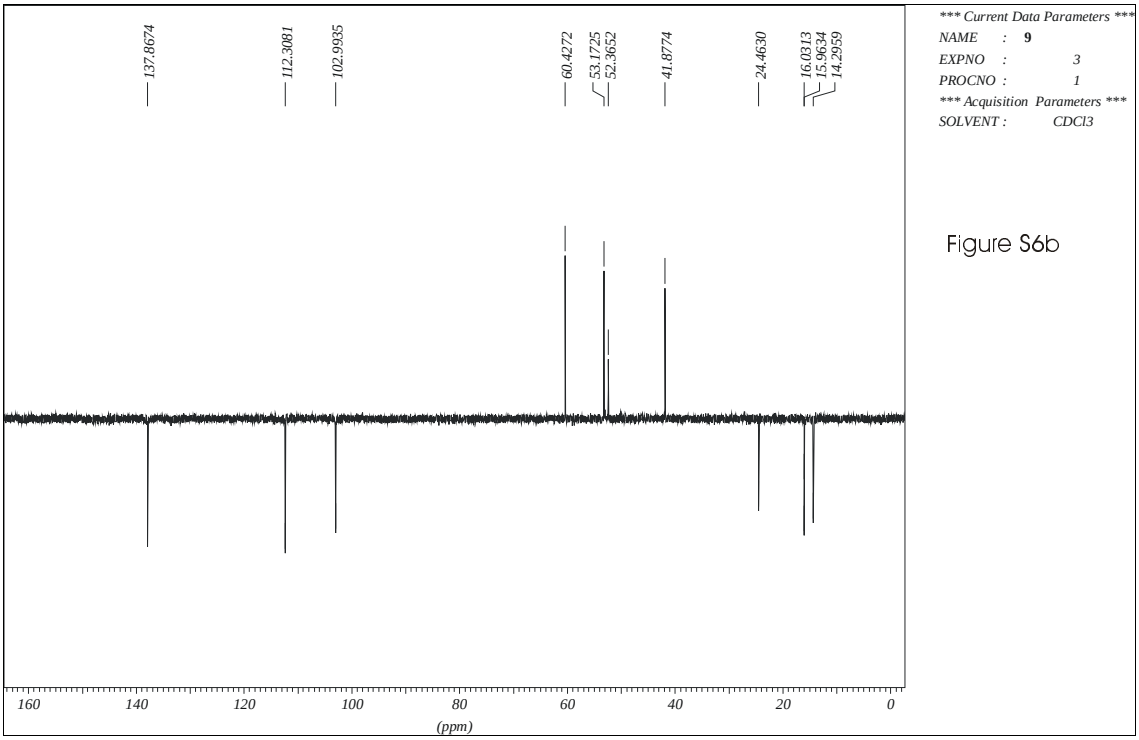


FIGURE S6. ^{13}C NMR spectra of **9** in $CDCl_3$.

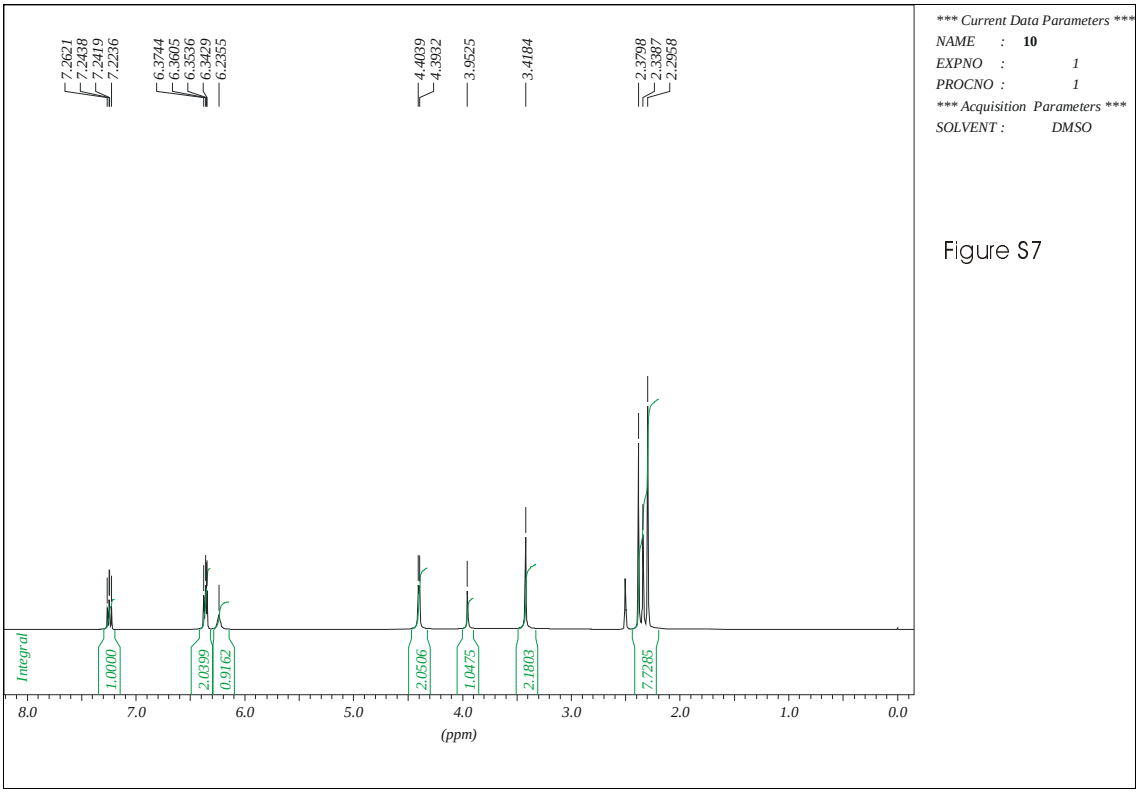


FIGURE S7. ^{1}H NMR spectrum of **10** in DMSO.

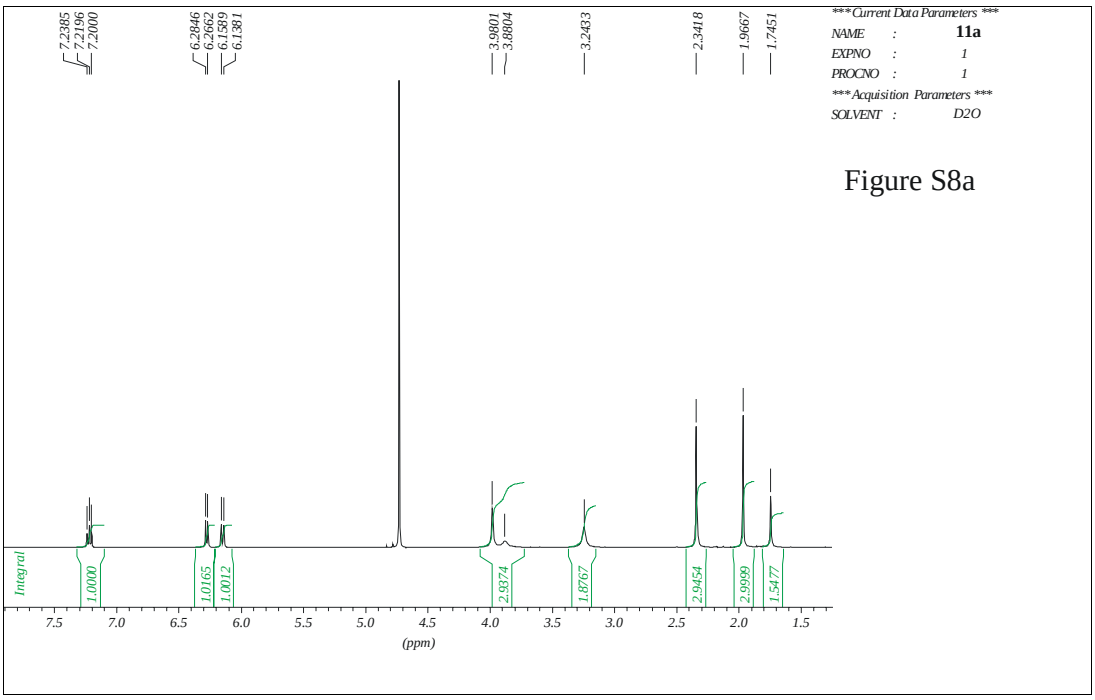


Figure S8a

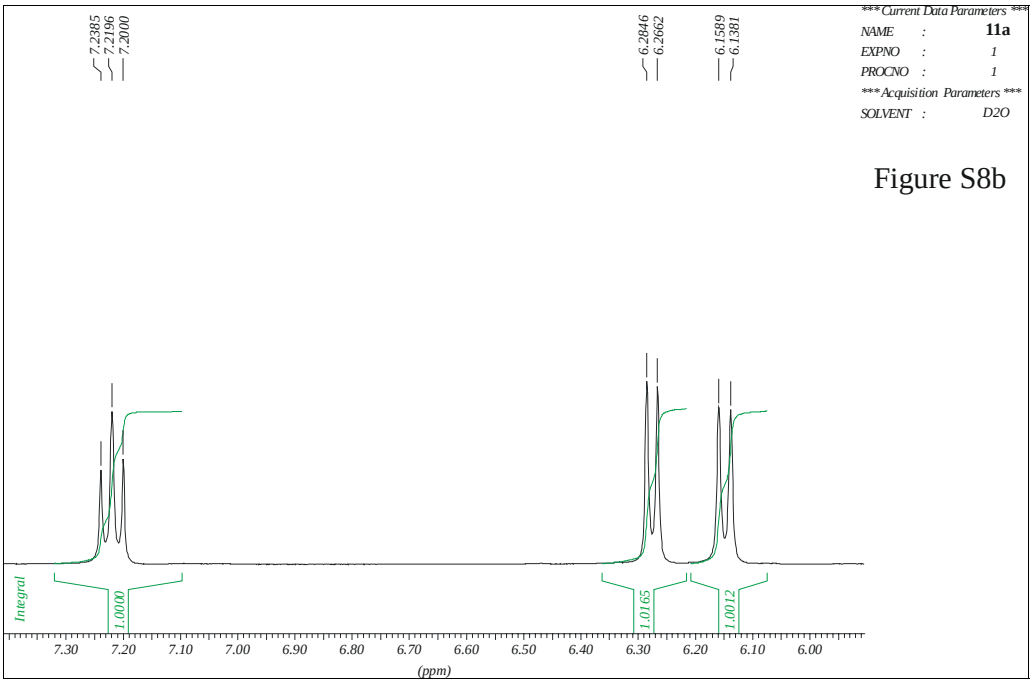


Figure S8b

FIGURE S8. ^{1}H NMR spectra of **11a** in D_2O ([**11a**] = 45 mM).

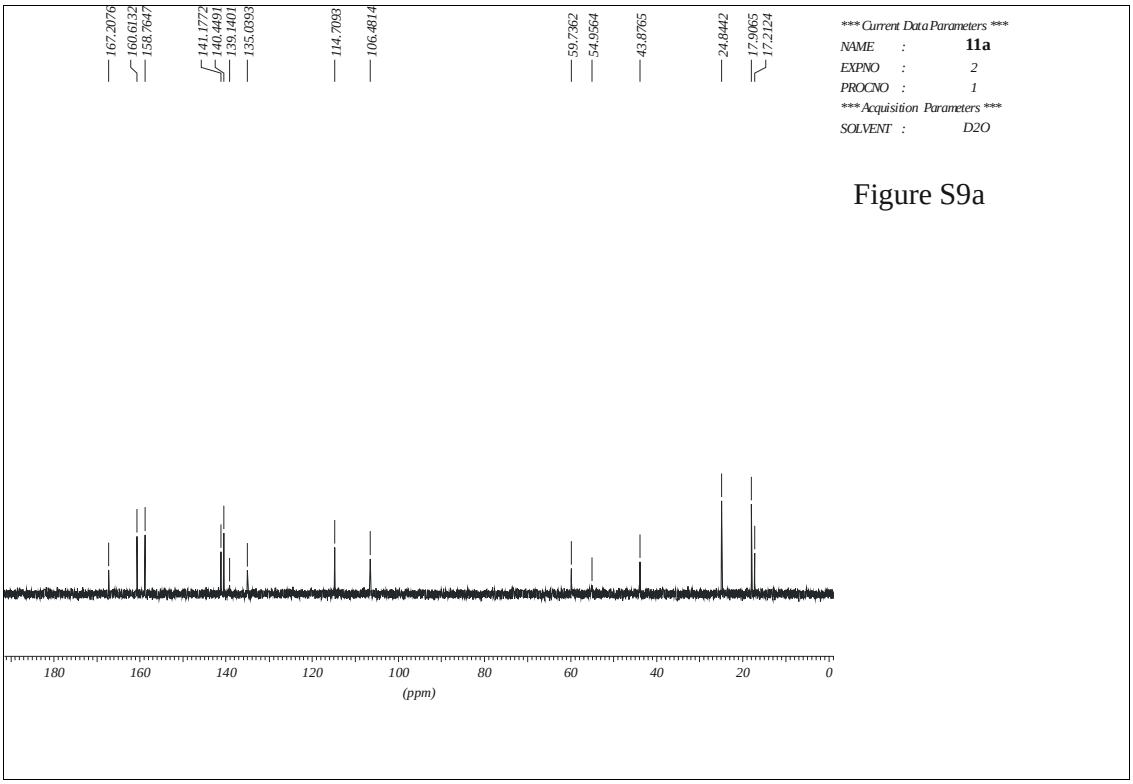


Figure S9a

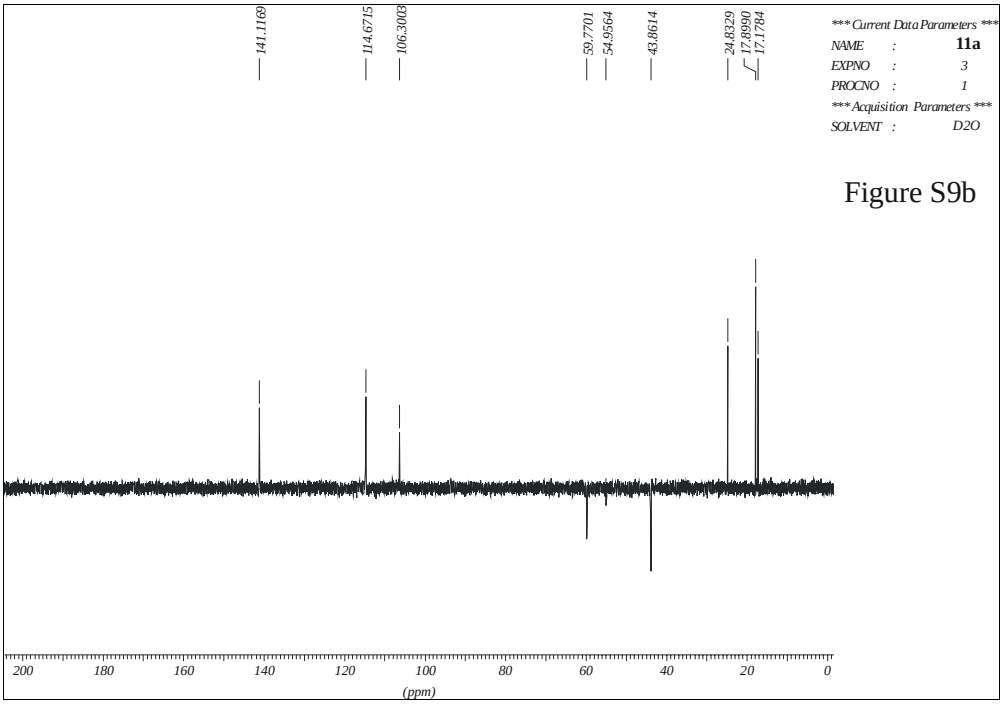


Figure S9b

FIGURE S9. ^{13}C NMR spectra of **11a** in D_2O ([**11a**] = 45 mM).

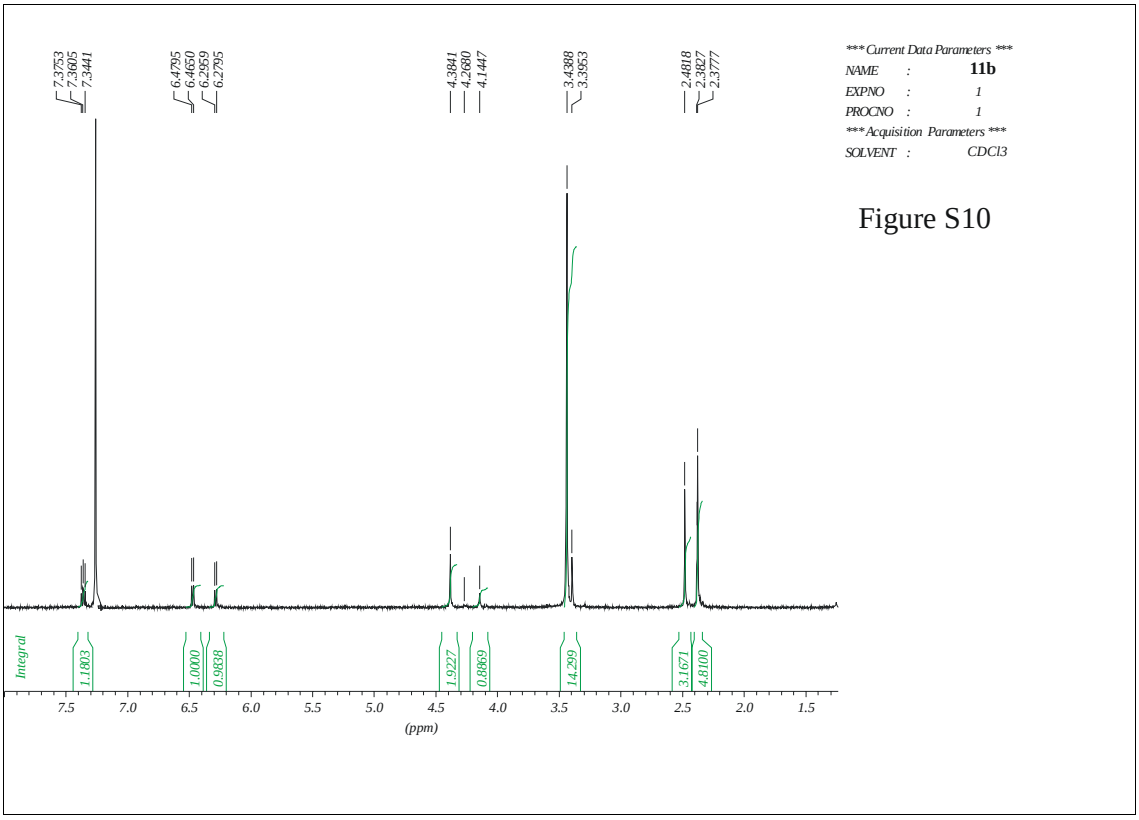


Figure S10

FIGURE S10. ^{1}H NMR spectrum of **11a** in $CDCl_3$ ([**11b**] = 0.85 mM).

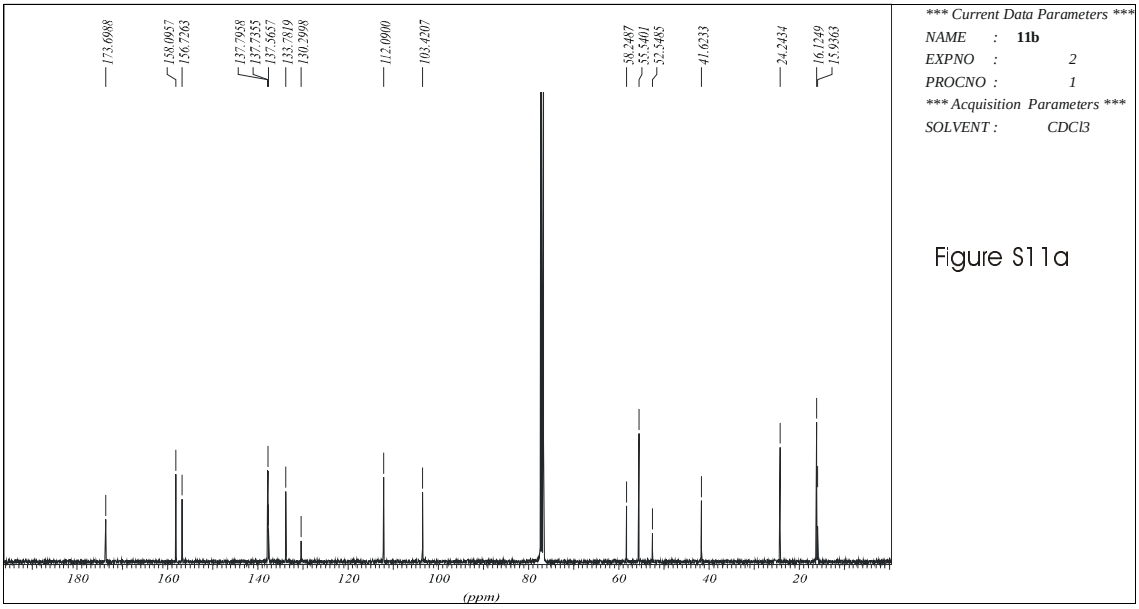


Figure S11a

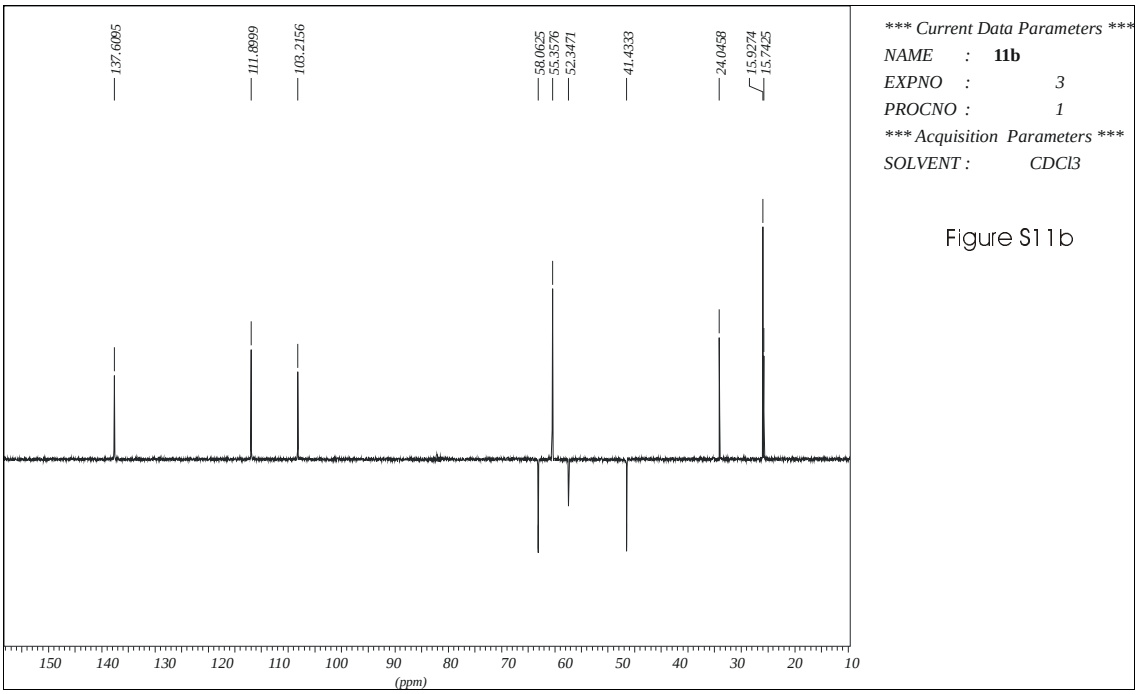


FIGURE S11. ^{13}C NMR spectra of **11b** in $CDCl_3$ ([**11b**] = 25 mM).

7. Crystal data for compound **9** (Tables of atomic coordinates and equivalent isotropic displacement parameters, bond lengths and angles, anisotropic displacement parameters, the anisotropic displacement factor, hydrogen coordinates and isotropic displacement parameters).

Table 1. Crystal data and structure refinement.

Identification code	majik	
Empirical formula	$C_{32}H_{43}N_5O_4$	
Formula weight	561.71	
Temperature	133(2) K	
Wavelength	0.71073 Å	
Crystal system	Monoclinic	
Space group	$P2_1/c$	
Unit cell dimensions	a = 11.6786(14) Å	□ = 90°
	b = 24.579(3) Å	□ = 95.713(5)°
	c = 10.7047(12) Å	□ = 90°
Volume	3057.5(6) Å^3	
Z	4	
Density (calculated)	1.220 Mg/m^3	
Absorption coefficient	0.081 mm^{-1}	
F(000)	1208	
Crystal size	0.35 x 0.15 x 0.15 mm^3	

Theta range for data collection	1.66 to 28.27°
Index ranges	-15<=h<=15, -32<=k<=32, -14<=l<=14
Reflections collected	30954
Independent reflections	7583 [R(int) = 0.0732]
Completeness to theta = 28.25°	99.8 %
Absorption correction	None
Refinement method	Full-matrix least-squares on F^2
Data / restraints / parameters	7583 / 0 / 385
Goodness-of-fit on F^2	0.938
Final R indices [I>2sigma(I)]	R1 = 0.0525, wR2 = 0.1222
R indices (all data)	R1 = 0.0998, wR2 = 0.1385
Largest diff. peak and hole	0.278 and -0.291 e.$Å^{-3}$

Table 2. Atomic coordinates (x 10^4) and equivalent isotropic displacement parameters ($Å^2$x 10^3). U(eq) is defined as one third of the trace of the orthogonalized U_{ij} tensor.

	x	y	z	U(eq)
C(1)	4689.6(14)	5363.6(7)	3209.4(15)	20.5(4)
C(2)	4841.1(15)	4874.0(7)	2571.9(16)	22.2(4)
C(3)	5650.9(15)	4848.6(7)	1682.4(16)	22.4(4)
C(4)	6327.6(15)	5301.5(7)	1464.5(15)	21.3(4)
C(5)	6155.4(14)	5793.8(7)	2086.6(15)	19.6(4)
C(6)	5331.1(15)	5829.7(7)	2946.4(15)	19.5(4)
C(7)	5128.5(16)	6359.8(7)	3611.2(18)	28.3(4)
C(8)	4148.2(17)	4366.6(8)	2815.2(19)	33.3(5)
C(9)	7264.3(17)	5261.8(8)	588.8(17)	31.9(5)
C(10)	6854.6(15)	6289.0(7)	1785.5(16)	23.8(4)
C(11)	3859.4(15)	5406.1(8)	4206.4(16)	25.3(4)
C(12)	5795.8(17)	4319.4(7)	979.4(18)	29.3(4)
C(13)	6859.6(15)	3449.3(7)	1368.2(16)	21.7(4)
C(14)	7773.5(16)	3165.5(8)	2030.2(17)	27.1(4)
C(15)	7931.2(17)	2631.2(8)	1745.5(18)	30.5(5)
C(16)	7214.5(16)	2389.9(7)	792.7(18)	29.2(4)
C(17)	6356.2(15)	2693.1(7)	157.4(17)	24.5(4)
C(18)	5573.9(17)	2451.7(8)	-901(2)	35.6(5)
C(19)	7059.1(15)	6854.6(7)	-93.2(16)	20.0(4)
C(20)	6638.7(16)	7014.3(7)	-1323.4(16)	25.1(4)
C(21)	7322.0(16)	7331.6(7)	-1980.3(17)	28.0(4)
C(22)	8395.6(16)	7501.9(7)	-1425.5(17)	26.2(4)
C(23)	8735.6(15)	7342.5(7)	-205.6(16)	22.1(4)

C(24)	9840.5(16)	7536.3(8)	496.4(18)	31.0(4)
C(25)	2156.9(15)	5493.7(7)	2653.1(16)	23.8(4)
C(26)	2133.0(16)	5833.3(7)	4817.6(17)	24.5(4)
C(27)	1394.6(16)	5917.3(7)	1974.0(17)	24.6(4)
C(28)	1718.5(15)	5372.6(7)	5605.7(17)	24.0(4)
C(29)	211.2(16)	6086.9(8)	79.0(18)	30.8(5)
C(30)	1244.1(18)	4438.3(8)	5736.0(19)	35.9(5)
C(31)	-283.0(18)	5771.4(9)	-1048.5(18)	38.6(5)
C(32)	1233(2)	3943.3(9)	4924(3)	57.2(7)
N(1)	6419.3(14)	6508.9(6)	571.1(14)	24.7(4)
N(2)	2799.7(12)	5703.3(6)	3783.1(13)	22.1(3)
N(3)	6639.4(14)	3976.9(6)	1700.9(15)	29.2(4)
N(4)	6163.5(12)	3222.4(6)	440.4(14)	23.0(3)
N(5)	8085.1(12)	7013.4(6)	453.0(13)	20.7(3)
O(1)	1148.7(13)	6354.4(5)	2375.5(13)	40.9(4)
O(2)	1488.9(12)	5432.8(6)	6669.2(12)	36.6(4)
O(3)	989.9(11)	5727.1(5)	840.3(11)	28.2(3)
O(4)	1606.7(11)	4895.4(5)	4999.9(12)	29.5(3)

Table 3. Bond lengths [Å] and angles [°].

C(1)-C(2)	1.403(2)	C(13)-C(14)	1.406(2)
C(1)-C(6)	1.412(2)	C(14)-C(15)	1.365(3)
C(1)-C(11)	1.515(2)	C(15)-C(16)	1.387(3)
C(2)-C(3)	1.409(3)	C(16)-C(17)	1.374(2)
C(2)-C(8)	1.523(2)	C(17)-N(4)	1.359(2)
C(3)-C(4)	1.398(2)	C(17)-C(18)	1.505(3)
C(3)-C(12)	1.520(2)	C(19)-N(5)	1.338(2)
C(4)-C(5)	1.405(2)	C(19)-N(1)	1.375(2)
C(4)-C(9)	1.513(3)	C(19)-C(20)	1.414(2)
C(5)-C(6)	1.399(2)	C(20)-C(21)	1.361(3)
C(5)-C(10)	1.518(2)	C(21)-C(22)	1.397(3)
C(6)-C(7)	1.515(2)	C(22)-C(23)	1.383(2)
C(10)-N(1)	1.452(2)	C(23)-N(5)	1.355(2)
C(11)-N(2)	1.469(2)	C(23)-C(24)	1.504(2)
C(12)-N(3)	1.458(2)	C(25)-N(2)	1.453(2)
C(13)-N(4)	1.341(2)	C(25)-C(27)	1.509(2)
C(13)-N(3)	1.376(2)	C(26)-N(2)	1.451(2)

C(26)-C(28)	1.520(2)	C(29)-O(3)	1.458(2)
C(27)-O(1)	1.203(2)	C(29)-C(31)	1.501(3)
C(27)-O(3)	1.342(2)	C(30)-O(4)	1.459(2)
C(28)-O(2)	1.204(2)	C(30)-C(32)	1.495(3)
C(28)-O(4)	1.340(2)		

C(2)-C(1)-C(6)	120.23(15)	C(16)-C(17)-C(18)	121.15(16)
C(2)-C(1)-C(11)	121.48(15)	N(5)-C(19)-N(1)	117.60(15)
C(6)-C(1)-C(11)	118.28(15)	N(5)-C(19)-C(20)	122.58(16)
C(1)-C(2)-C(3)	119.50(15)	N(1)-C(19)-C(20)	119.79(16)
C(1)-C(2)-C(8)	121.61(16)	C(21)-C(20)-C(19)	118.24(17)
C(3)-C(2)-C(8)	118.89(16)	C(20)-C(21)-C(22)	119.98(17)
C(4)-C(3)-C(2)	120.41(16)	C(23)-C(22)-C(21)	118.54(17)
C(4)-C(3)-C(12)	120.39(16)	N(5)-C(23)-C(22)	122.49(16)
C(2)-C(3)-C(12)	119.20(16)	N(5)-C(23)-C(24)	115.26(15)
C(3)-C(4)-C(5)	119.77(16)	C(22)-C(23)-C(24)	122.22(16)
C(3)-C(4)-C(9)	120.51(16)	N(2)-C(25)-C(27)	112.81(14)
C(5)-C(4)-C(9)	119.70(16)	N(2)-C(26)-C(28)	118.96(15)
C(6)-C(5)-C(4)	120.42(16)	O(1)-C(27)-O(3)	124.01(17)
C(6)-C(5)-C(10)	120.59(15)	O(1)-C(27)-C(25)	126.50(17)
C(4)-C(5)-C(10)	118.97(15)	O(3)-C(27)-C(25)	109.47(15)
C(5)-C(6)-C(1)	119.55(15)	O(2)-C(28)-O(4)	123.01(17)
C(5)-C(6)-C(7)	120.88(15)	O(2)-C(28)-C(26)	123.11(17)
C(1)-C(6)-C(7)	119.56(15)	O(4)-C(28)-C(26)	113.86(15)
N(1)-C(10)-C(5)	109.93(14)	O(3)-C(29)-C(31)	107.73(16)
N(2)-C(11)-C(1)	113.29(14)	O(4)-C(30)-C(32)	107.40(17)
N(3)-C(12)-C(3)	109.54(14)	C(19)-N(1)-C(10)	122.21(15)
N(4)-C(13)-N(3)	117.95(15)	C(26)-N(2)-C(25)	116.18(14)
N(4)-C(13)-C(14)	122.73(16)	C(26)-N(2)-C(11)	111.93(14)
N(3)-C(13)-C(14)	119.29(16)	C(25)-N(2)-C(11)	115.46(14)
C(15)-C(14)-C(13)	118.52(17)	C(13)-N(3)-C(12)	122.76(15)
C(14)-C(15)-C(16)	119.40(17)	C(13)-N(4)-C(17)	117.49(15)
C(17)-C(16)-C(15)	119.29(17)	C(19)-N(5)-C(23)	118.10(15)
N(4)-C(17)-C(16)	122.53(16)	C(27)-O(3)-C(29)	115.99(14)
N(4)-C(17)-C(18)	116.32(16)	C(28)-O(4)-C(30)	115.60(14)

Table 4. Torsion angles [°].

C(6)-C(1)-C(2)-C(3)	1.0(2)	N(1)-C(19)-C(20)-C(21)	-176.33(16)
C(11)-C(1)-C(2)-C(3)	-177.86(15)	C(19)-C(20)-C(21)-C(22)	-1.6(3)
C(6)-C(1)-C(2)-C(8)	-178.88(15)	C(20)-C(21)-C(22)-C(23)	-0.2(3)
C(11)-C(1)-C(2)-C(8)	2.3(2)	C(21)-C(22)-C(23)-N(5)	2.3(3)
C(1)-C(2)-C(3)-C(4)	2.1(2)	C(21)-C(22)-C(23)-C(24)	-175.89(17)
C(8)-C(2)-C(3)-C(4)	-177.96(16)	N(2)-C(25)-C(27)-O(1)	-12.7(3)
C(1)-C(2)-C(3)-C(12)	-178.97(15)	N(2)-C(25)-C(27)-O(3)	169.12(14)
C(8)-C(2)-C(3)-C(12)	0.9(2)	N(2)-C(26)-C(28)-O(2)	156.22(17)
C(2)-C(3)-C(4)-C(5)	-3.5(2)	N(2)-C(26)-C(28)-O(4)	-25.2(2)
C(12)-C(3)-C(4)-C(5)	177.67(15)	N(5)-C(19)-N(1)-C(10)	-4.2(2)
C(2)-C(3)-C(4)-C(9)	175.00(16)	C(20)-C(19)-N(1)-C(10)	173.83(16)
C(12)-C(3)-C(4)-C(9)	-3.9(2)	C(5)-C(10)-N(1)-C(19)	-161.61(16)
C(3)-C(4)-C(5)-C(6)	1.6(2)	C(28)-C(26)-N(2)-C(25)	76.6(2)
C(9)-C(4)-C(5)-C(6)	-176.85(15)	C(28)-C(26)-N(2)-C(11)	-59.0(2)
C(3)-C(4)-C(5)-C(10)	-176.77(15)	C(27)-C(25)-N(2)-C(26)	69.4(2)
C(9)-C(4)-C(5)-C(10)	4.8(2)	C(27)-C(25)-N(2)-C(11)	-156.56(15)
C(4)-C(5)-C(6)-C(1)	1.5(2)	C(1)-C(11)-N(2)-C(26)	-168.33(14)
C(10)-C(5)-C(6)-C(1)	179.88(15)	C(1)-C(11)-N(2)-C(25)	55.7(2)
C(4)-C(5)-C(6)-C(7)	-179.29(16)	N(4)-C(13)-N(3)-C(12)	-8.9(3)
C(10)-C(5)-C(6)-C(7)	-0.9(2)	C(14)-C(13)-N(3)-C(12)	172.99(18)
C(2)-C(1)-C(6)-C(5)	-2.8(2)	C(3)-C(12)-N(3)-C(13)	172.81(17)
C(11)-C(1)-C(6)-C(5)	176.08(15)	N(3)-C(13)-N(4)-C(17)	-177.07(16)
C(2)-C(1)-C(6)-C(7)	177.96(16)	C(14)-C(13)-N(4)-C(17)	1.0(3)
C(11)-C(1)-C(6)-C(7)	-3.1(2)	C(16)-C(17)-N(4)-C(13)	0.9(3)
C(6)-C(5)-C(10)-N(1)	-103.54(18)	C(18)-C(17)-N(4)-C(13)	-179.06(16)
C(4)-C(5)-C(10)-N(1)	74.9(2)	N(1)-C(19)-N(5)-C(23)	178.31(15)
C(2)-C(1)-C(11)-N(2)	-105.78(18)	C(20)-C(19)-N(5)-C(23)	0.4(2)
C(6)-C(1)-C(11)-N(2)	75.3(2)	C(22)-C(23)-N(5)-C(19)	-2.3(2)
C(4)-C(3)-C(12)-N(3)	91.1(2)	C(24)-C(23)-N(5)-C(19)	175.96(15)
C(2)-C(3)-C(12)-N(3)	-87.8(2)	O(1)-C(27)-O(3)-C(29)	0.3(3)
N(4)-C(13)-C(14)-C(15)	-2.4(3)	C(25)-C(27)-O(3)-C(29)	178.49(15)
N(3)-C(13)-C(14)-C(15)	175.59(18)	C(31)-C(29)-O(3)-C(27)	-171.37(16)
C(13)-C(14)-C(15)-C(16)	2.0(3)	O(2)-C(28)-O(4)-C(30)	-3.2(3)
C(14)-C(15)-C(16)-C(17)	-0.3(3)	C(26)-C(28)-O(4)-C(30)	178.20(15)
C(15)-C(16)-C(17)-N(4)	-1.2(3)	C(32)-C(30)-O(4)-C(28)	-176.99(17)
C(15)-C(16)-C(17)-C(18)	178.70(18)		
N(5)-C(19)-C(20)-C(21)	1.6(3)		

Table 5. Hydrogen bonds [Å and °].

D-H...A	d(D-H)	d(H...A)	d(D...A)	<(DHA)
N(1)-H(01)...N(4)#1	0.84(2)	2.36(2)	3.171(2)	162.4(18)
N(3)-H(03)...O(2)#2	0.873(19)	2.16(2)	3.028(2)	176.9(18)

Symmetry transformations used to generate equivalent atoms:

#1 -x+1,-y+1,-z

#2 -x+1,-y+1,-z+1