Supporting Information

Using "small" molecules to facilitate exchange of bicarbonate and chloride anions across liposomal membranes.

Jeffery T. Davis,[1] Philip A. Gale,[2] O.A. Okunola,[1] Pilar Prados,[3] Jose Carlos Iglesias-Sánchez,[3] Tomás Torroba,[4] and Roberto Quesada[4]

[1]Department of Chemistry and Biochemistry, University of Maryland, College Park, MD 20742, USA. [2]School of Chemistry, University of Southampton, Southampton, SO17 1BJ, United Kingdom. [3]Departamento de Química Orgánica, Universidad Autónoma de Madrid, 28049 Madrid, Spain. [4]Departamento de Química, Universidad de Burgos, 09001 Burgos, Spain. Correspondence should be addressed to J.T.D. (jdavis@umd.edu), P.A.G. (philip.gale@soton.ac.uk) or R.Q. (rquesada@ubu.es)

Experimental procedures

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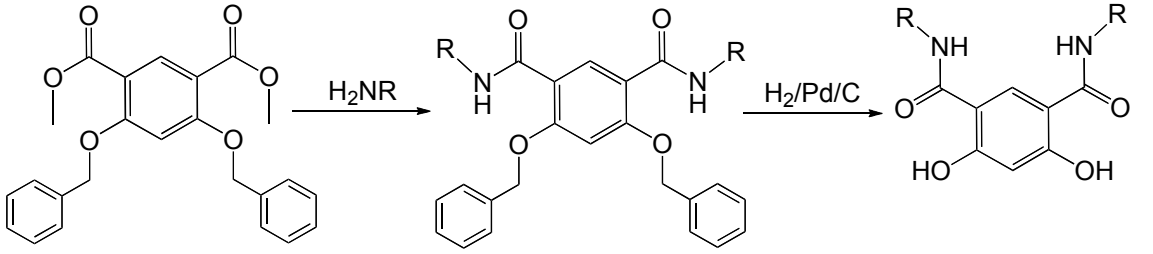
General Methods. Unless otherwise reported, all reactions were carried out under dry and deoxygenated argon atmosphere. Solvents were freshly distilled and dried before use by standard methods. All chemicals were used as purchased. Reported melting points are uncorrected and were measured in open capillaries on a Gallenkamp Melting Point apparatus. The NMR experiments (^{1}H, $^{13}C\{1H\}$) were carried out at 500 (125) MHz or 300 (75) MHz and reported chemical shifts (δ) are externally referenced to solvent residual signal and given in ppm. Mass spectra were performed on a REFLEX spectrometer. Elemental analyses, performed on a LECO CHN 932 microanalyser. Prodigiosin was a gift of the National Cancer Institute at NIH. Compound **2** was reported previously by us.[1] Synthesis outline:



8

5 R = n-Bu
6 R = 3-methylbutyl
7 R = n-Oct

2 R = n-Bu
3 R = 3-methylbutyl
4 R = n-Oct

Dimethyl-4,6-(dibenzyloxy)isophthalate 8. To a suspension of dimethyl 4,6-dihydroxyisophthalate[2] (500 mg, 1.9 mmol) and K_2CO_3 (1.3 gr, 9.6 mmol) in acetone (100 mL), benzyl bromide (1.1 mL, 9.6 mmol) was added and the mixture was heated under reflux for 12 h. The mixture was cooled to room temperature and quenched with 1M HCl, stirred for 30 min and extracted with CH_2Cl_2. The organic layer was washed with H_2O, dried ($MgSO_4$) and evaporated

[1] Santacroce, P. V. *et al.* Conformational Control of Transmembrane Cl^- Transport *J. Am. Chem. Soc.* **129**, 1886-1887 (2007).
[2] Zeng, H., Miller, R.; Flowers, R. A.; Gong, B. A Highly Stable, Six-Hydrogen-Bonded Molecular Duplex, *J. Am. Chem. Soc.* ***122***, 2635-2644 (2000).

to give compound **8** (703 mg, 91%) as a white solid. mp: 145 ºC; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 3.19 (OCH_3), 5.17 (CH_2Ar), 6.55 (s, 1H, ArH), 7.33-7.47 (m, 10H, ArH), 8.52 (s, 1H, ArH); ^{13}C NMR (125 MHz, $CDCl_3$, DEPT): δ = 51.9 (OCH_3), 70.7 (CH_2Ar), 99.4, 112.5 (ArH), 126.7 (Ar), 128.1, 128.7 (ArH), 135.9, 137.1 (Ar), 162.2 (CO); ESI-MS (positive ion) *m/z*: 406.4 [M^+]; analysis (calcd., found for $C_{24}H_{22}O_6$): C (70.92, 70.88), H (5.46, 5.41).

4,6-Dibenzyloxy-*N,N'*-dibutylisophthalamide 5 A solution of dimethyl 4,6-(dibenzyloxy)isophtalate **8** (450 mg, 1.1 mmol) in DMF (5 mL) was added dropwise to a solution of 1-butylamine (4 eq in 2 mL of DMF). The mixture was heated at 110 ºC in a sealed tube for 24 h. The reaction mixture was poured into 1M HCl, and the precipitated material was filtered and washed with water and dried, to give **5** as a white solid (434 mg, 81%). mp: 132 ºC; ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.72 (m, 6H, CH_3), 1.07 (m, 4H, CH_2), 1.24 (m, 4H, CH_2), 3.24 (m, 4H, CH_2NH), 5.07 (s, 4H, CH_2Ar), 6.54 (s,1H, ArH), 7.34 (m, 12H, ArH and NH), 8.93 (s, 1H, ArH); ^{13}C NMR (125 MHz, $CDCl_3$, DEPT): δ = 14.1 (CH_3), 20.9, 32.6 (CH_2), 40.0 (CH_2NH), 70.1 (CH_2Ar), 96.9 (ArH), 115.9 (Ar), 127.0, 127.8, 128.6 (ArH), 136.2 (Ar), 136.4 (ArH), 163.5 (Ar), 165.0 (CO); ESI-MS (positive ion) *m/z*: 489.3 [M^+]. analysis (calcd., found for $C_{30}H_{36}N_2O_4$): C (73.74, 73.71), H (7.43, 7.40) N (5.73, 5.68).

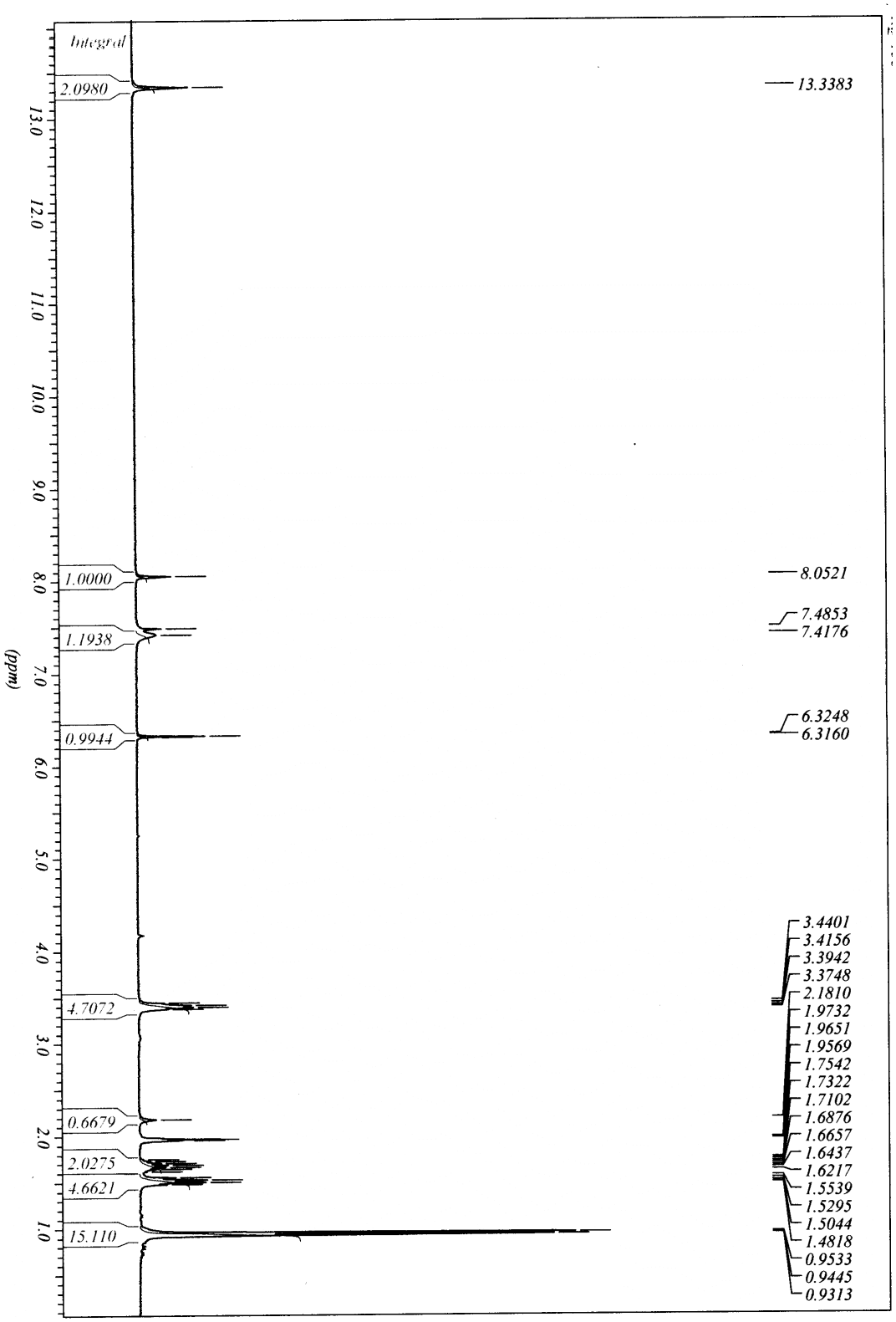
4,6-Dibenzyloxy-*N,N'*-diisopentylisophthalamide 6. This compound was prepared similarly to **5** from **8** (450 mg, 1.1 mmol) and isoamylamine, to give **6** (237 mg, 64%). ^{1}H NMR (500 MHz, d_6-DMSO): δ = 0.90 (d, J = 6.0 Hz, 12H, CH_3), 1.39 (m, 4H, CH_2), 1.73 (m, 2H, CH), 3.33 (m, 4H, CH_2NH), 5.22 (s, 4H, CH_2Ar), 6.63 (s, 1H, ArH), 7.43 (br, 10H, ArH), 7.52 (br, 2H, NH), 9.03 (s, 1H, ArH); ^{13}C NMR (125 MHz, d_6-DMSO, DEPT): δ = 21.9 (CH_3), 25.4 (CH), 38.5 (CH_2NH), 53.0 (CH_2), 97.4, 115.9, 120.1, 129.6, 137.0 (ArH), 136.9, 159.4, (Ar), 161.8, 164.1 (CO); ESI-MS (positive ion) *m/z*: 517.7 [M^+]; analysis (calcd., found for $C_{32}H_{40}N_2O_4$): C (74.39, 74.59), H (7.80, 7.69) N (5.42, 5.62).

4,6-Dibenzyloxy-*N,N'*-dioctylisophthalamide 7. This compound was prepared similarly to **5** from **8** and 1-octylamine to give **7** (490 mg, 74%). mp 138 ºC; ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.91 (t, J = 7.3 Hz, 6H, CH_3), 1.28-1.34 (m, 24H, CH_2), 3.33 (m, 4H, CH_2NH), 5.18 (s, 4H, CH_2Ar), 6.63 (s, 1H, ArH), 7.45 (bs, 10H, ArH), 7.52 (bs, 2H, NH), 9.03 (s, 1H, ArH); ^{13}C NMR (125 MHz, $CDCl_3$, DEPT): δ = 14.1 (CH_3), 22.7, 27.0, 29.2, 29.22, 29.3, 31.9 (CH_2), 39.8 (CH_2NH), 71.7 (OCH_2Ar), 97.4 (ArH), 115.9 (Ar), 121.1, 129.0, 135.0 (ArH), 136.9, 159.4, (Ar), 164.1 (CO); ESI-MS (positive ion) *m/z*: 601.8 [M^+]. analysis (calcd., found for $C_{38}H_{52}N_2O_4$): C (75.96, 75.91), H (8.72, 8.69), N (4.66, 4.62).

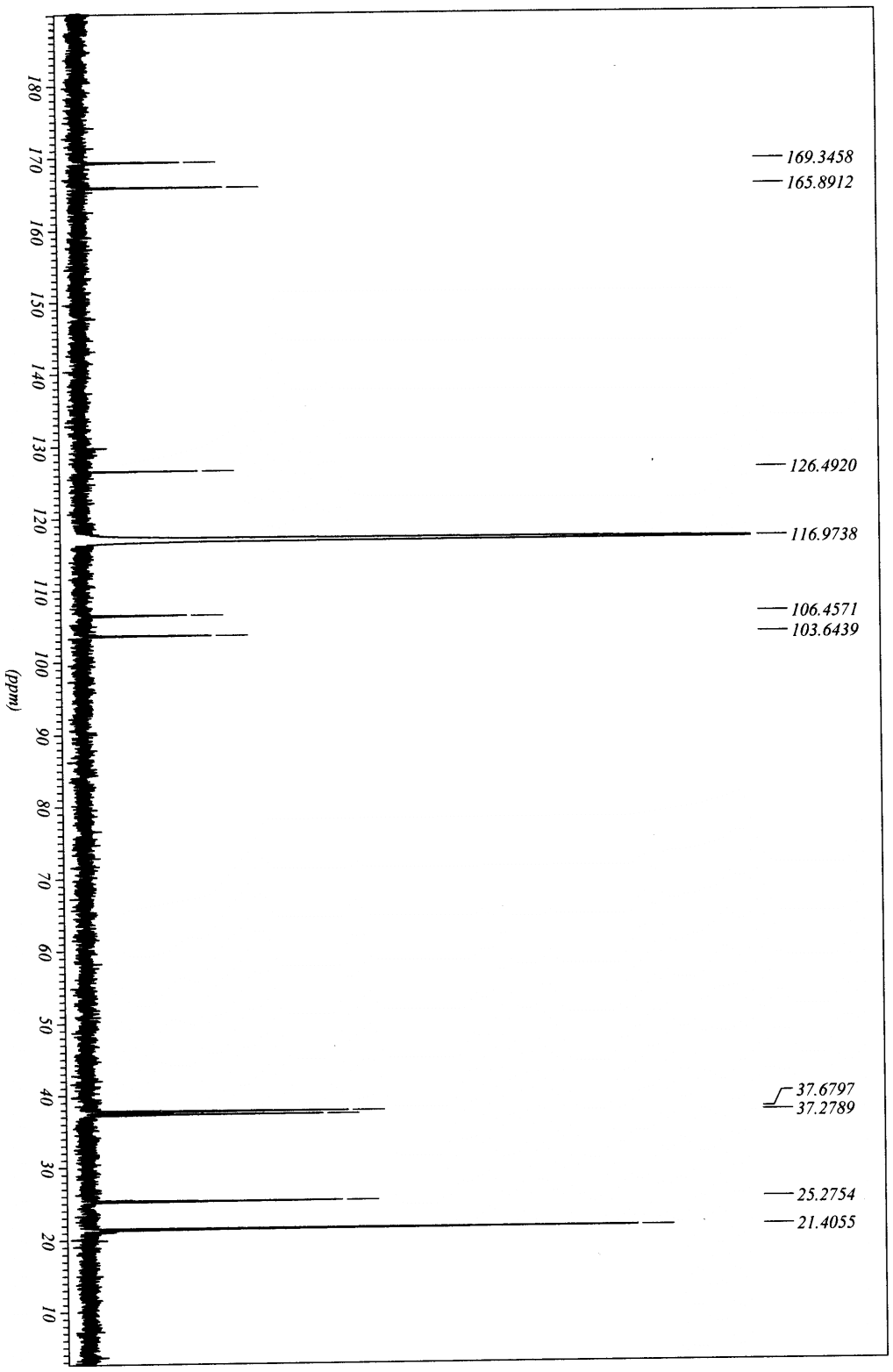
***N,N'*-dibutyl-4,6-dihydroxyisophthalamide 2.** 200 mg (0.41 mmol) of 4,6-dibenzyloxy-*N,N'*-dibutylisophthalamide **5** were dissolved in a THF (20 mL)/ MeOH (20 mL) mixture and 10 % Pd/C (10 % eq) was added. Through this suspension hydrogen was bubbled during 30 min. The mixture was filtered on Celite, and the solvent was evaporated to give a **2** as a white solid (quantitative). mp 170 ºC; ^{1}H NMR (CD_3CN): δ = 0.96 (t, 6H, J = 7.3 Hz, CH_3), 1.44 (m, 4H, CH_2), 1.60 (m, 4H, CH_2), 3.39 (m, 4H, CH_2NH), 6.32 (s, 1H, ArH), 7.11 (bs, 2H, NH), 7.80 (s, 1H, AH), 13.25 (s, 2H, OH); ^{13}C NMR (CD_3CN): δ = 12.8 (CH_3), 19.5, 30.9 (CH_2), 38.7 (CH_2NH), 103.7 (ArH), 106.5 (Ar), 126.1 (ArH), 165.8 (Ar), 169.3 (CO); ESI-MS (positive ion): m/z: 309.2 [M+]; analysis (calcd., found for $C_{16}H_{24}N_2O_4$): C (62.32, 62.71), H (7.84,7.94), N (9.08; 8.92).

***N,N'*-diisopentyl-4,6-dihydroxyisophthalamide 3.** This compound was prepared using the same procedure as described above from **6** to give **3** (quantitative). ^{1}H NMR (300 MHz, CD_3CN): δ = 0.94 (d, 12H, J = 7.3 Hz, CH_3), 1.51 (m, 4H, CH_2), 1.67 (m, 2H, CH), 3.40 (m, 4H, CH_2NH), 6.31 (s, 1H, ArH), 7.41 (br, 2H, NH), 8.05 (s, 1H, ArH), 13.34 (s, 2H, OH); ^{13}C NMR (125 MHz, CD_3CN, DEPT): δ = 21.4 (CH_3), 25.3 (CH), 37.3 (CH_2), 37.7 (CH_2), 103.6 (ArH), 106.5 (Ar), 126.5 (ArH), 165.9 (Ar), 169.3 (CO); ESI-MS (positive ion) *m/z*: 337.2 [M^+]; analysis (calcd., found for $C_{18}H_{28}N_2O_4$):C (64.26, 64.44), H (8.39, 8.51), N (8.33; 8.30).

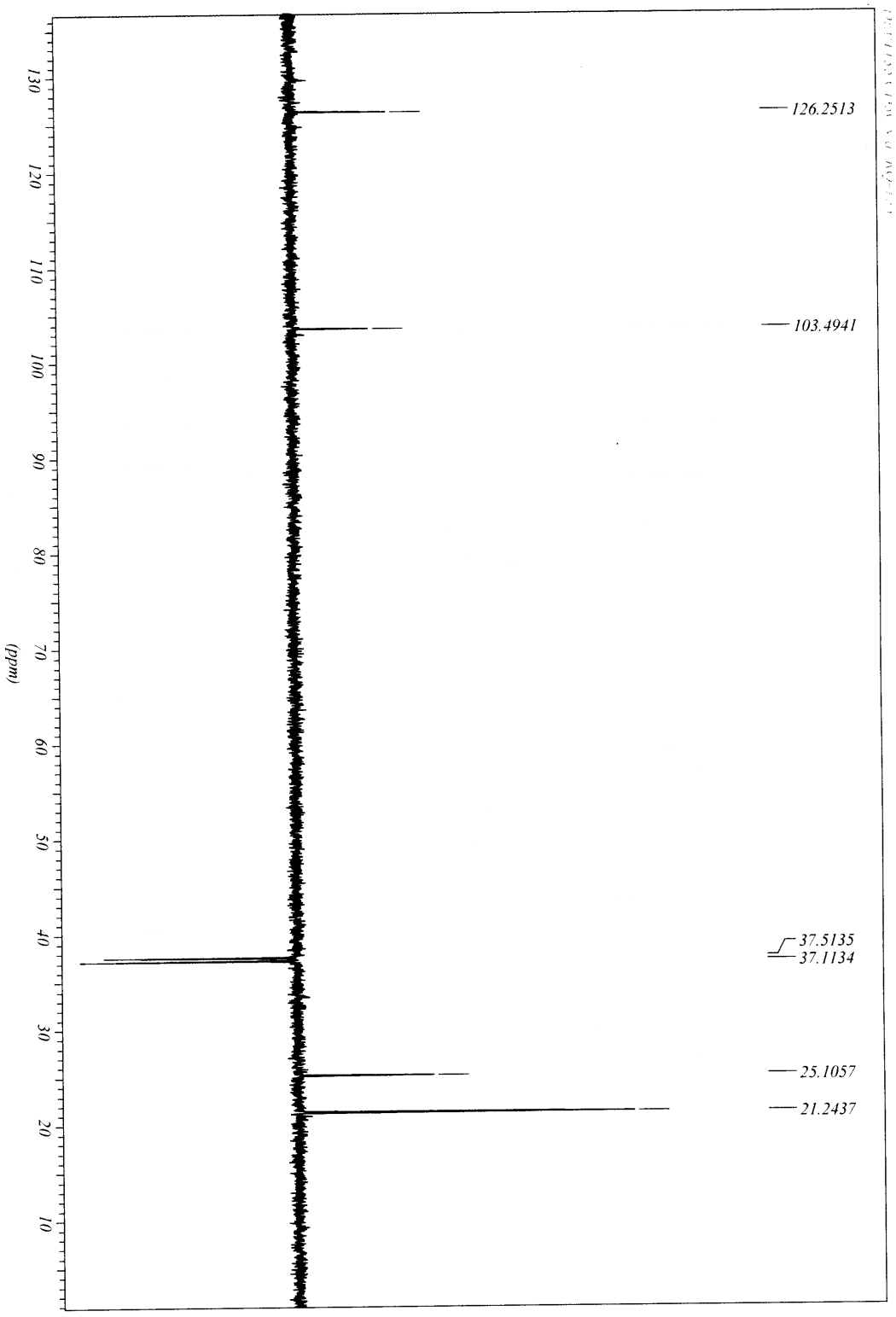
***N,N'*-dioctyl-4,6-dihydroxyisophthalamide 4.** As described above, this compound was prepared from **7** to give **4** (quantitative). mp 150 ºC*;* ^{1}H NMR (500 MHz, $CDCl_3$): δ = 0.78 (m, 6H, CH_3), 1.15, (m, 16H, CH_2), 1.44 (m, 4H, CH_2), 3.24 (m, 4H, CH_2NH), 6.41 (s, 1H, ArH), 6.94 (br, 2H, NH), 7.89 (s, 1H, ArH), 12.73 (s, 2H, OH); ^{13}C NMR (125 MHz, $CDCl_3$, DEPT): δ = 14.1 (CH_3), 22.6, 27.0, 29.2, 29.4, 31.8 (CH_2), 40.0 (CH_2NH), 105.1 (ArH), 107.6 (Ar), 126.4 (ArH), 165.3 (Ar), 169.0 (CO); ESI-MS (positive ion) *m/z*: 421.3 [M^+]. analysis (calcd., found for $C_{24}H_{40}N_2O_4$): C 68.54, 68.41), H (9.59, 9.57), N (6.66; 6.30).



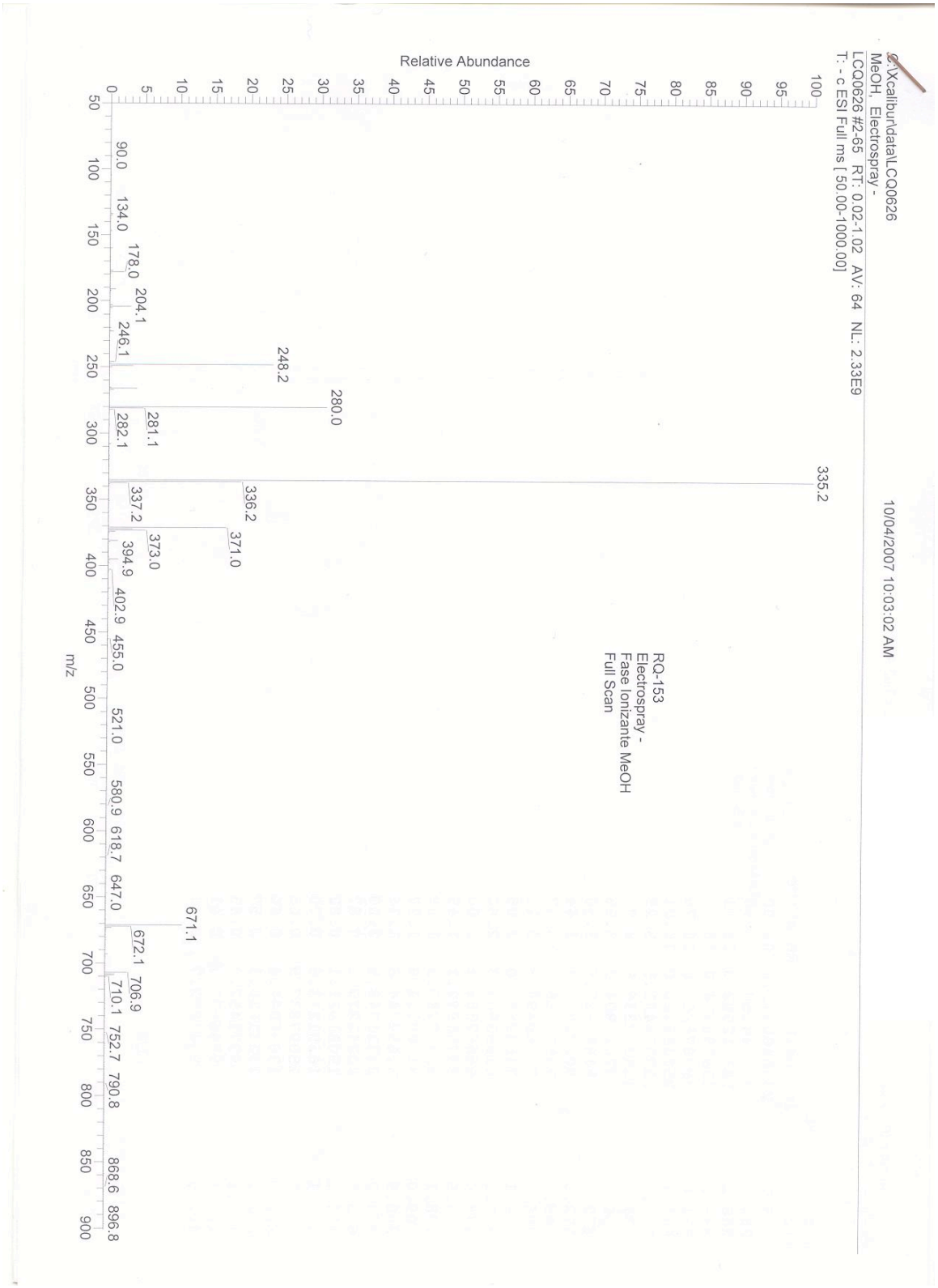
Supplementary figure S1: ^{1}H NMR (CD_3CN) spectrum of **3**.



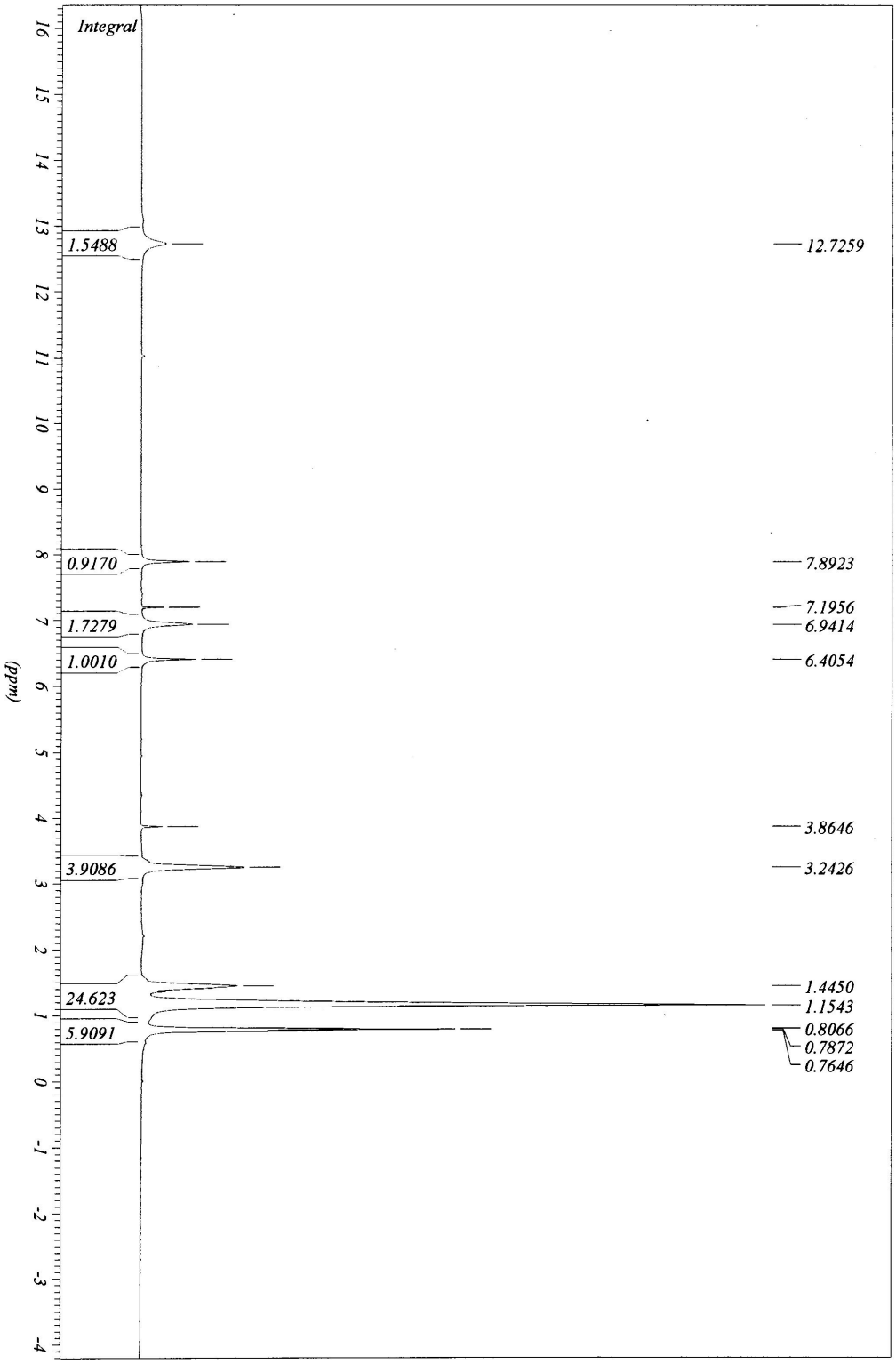
Supplementary figure S2: ^{13}C NMR (CD_3CN) spectrum of **3**.



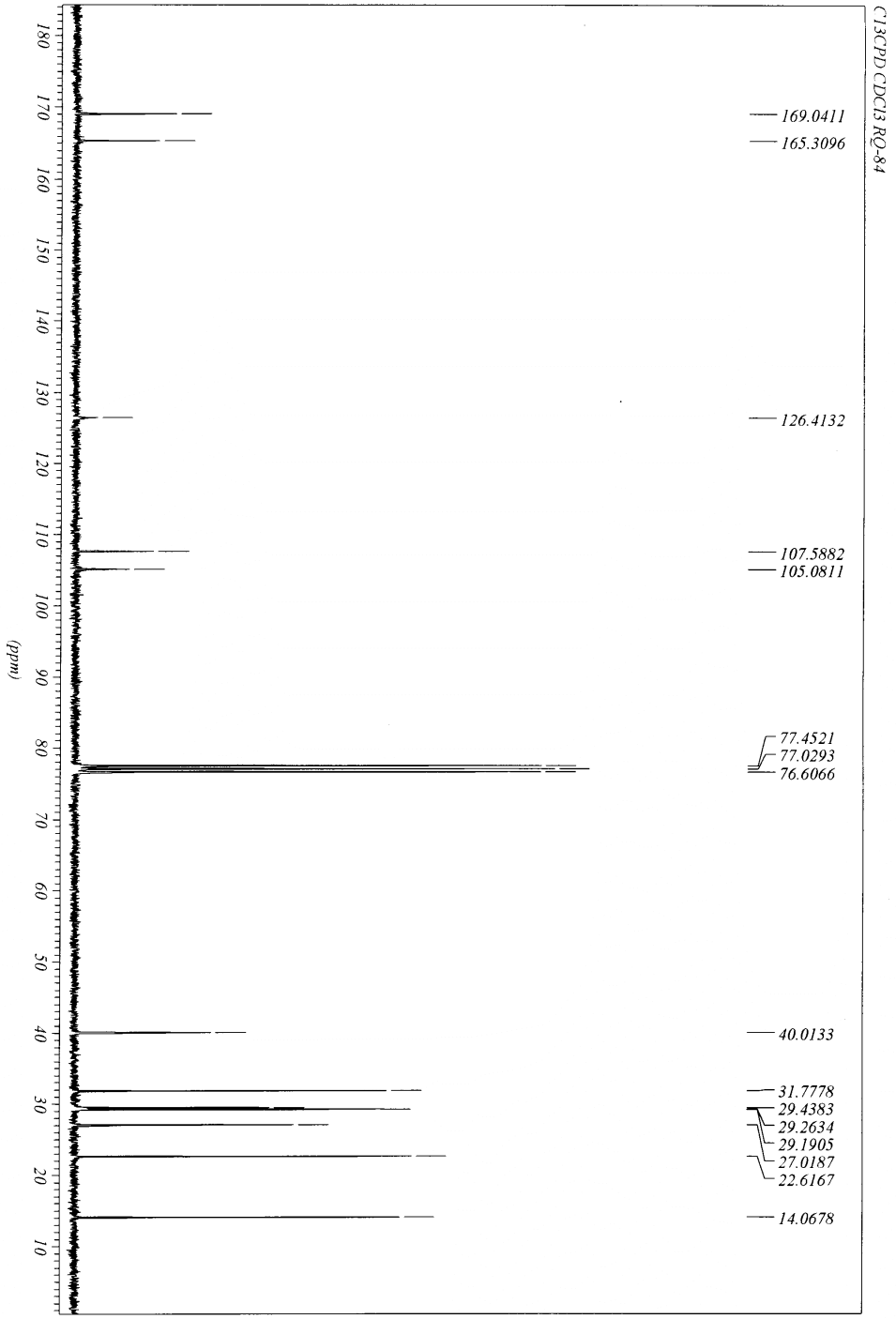
Supplementary figure S3: ^{13}C NMR DEPT (CD_3CN) spectrum of **3**.



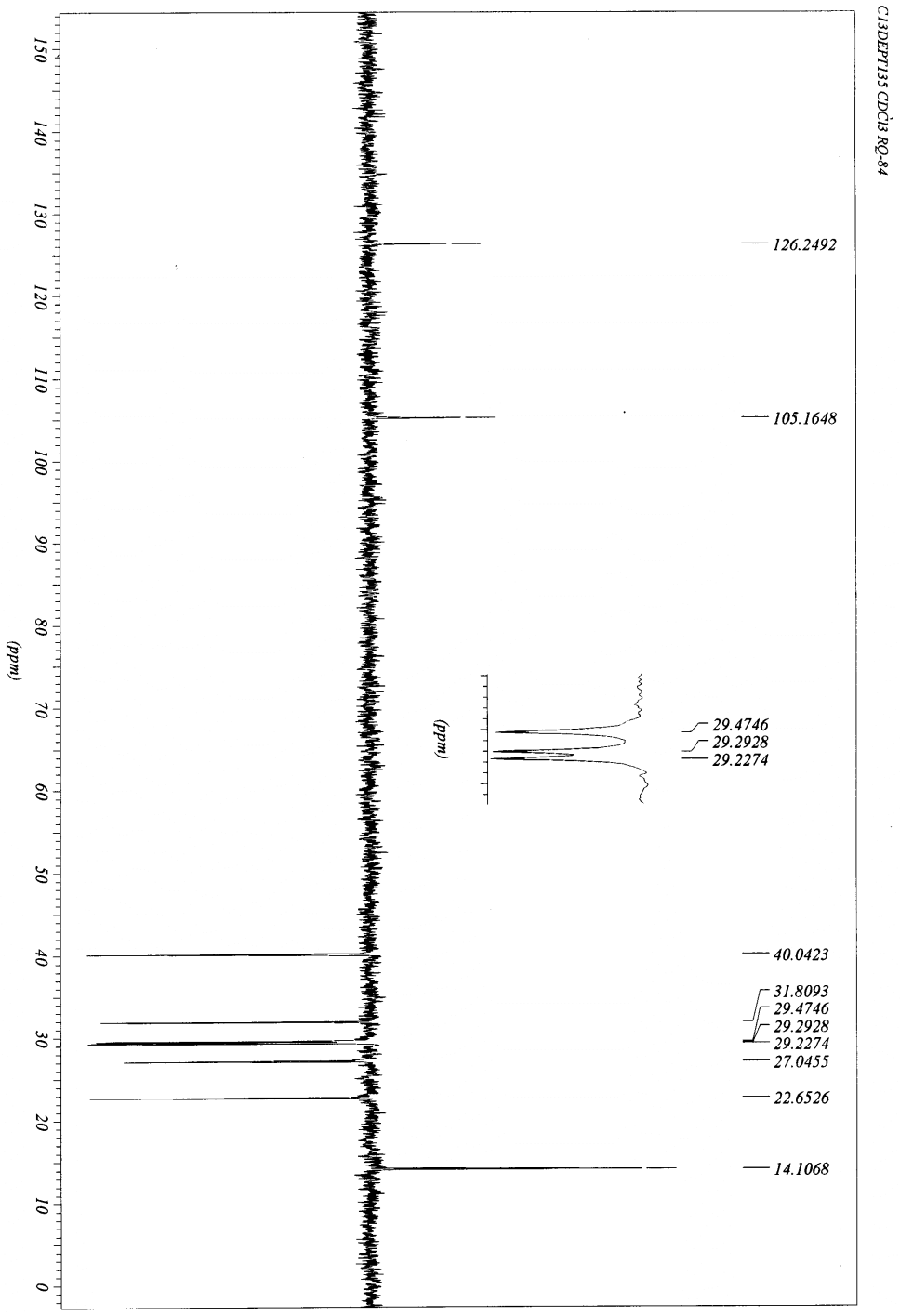
Supplementary figure S4: ESI mass spectrum of **3**.



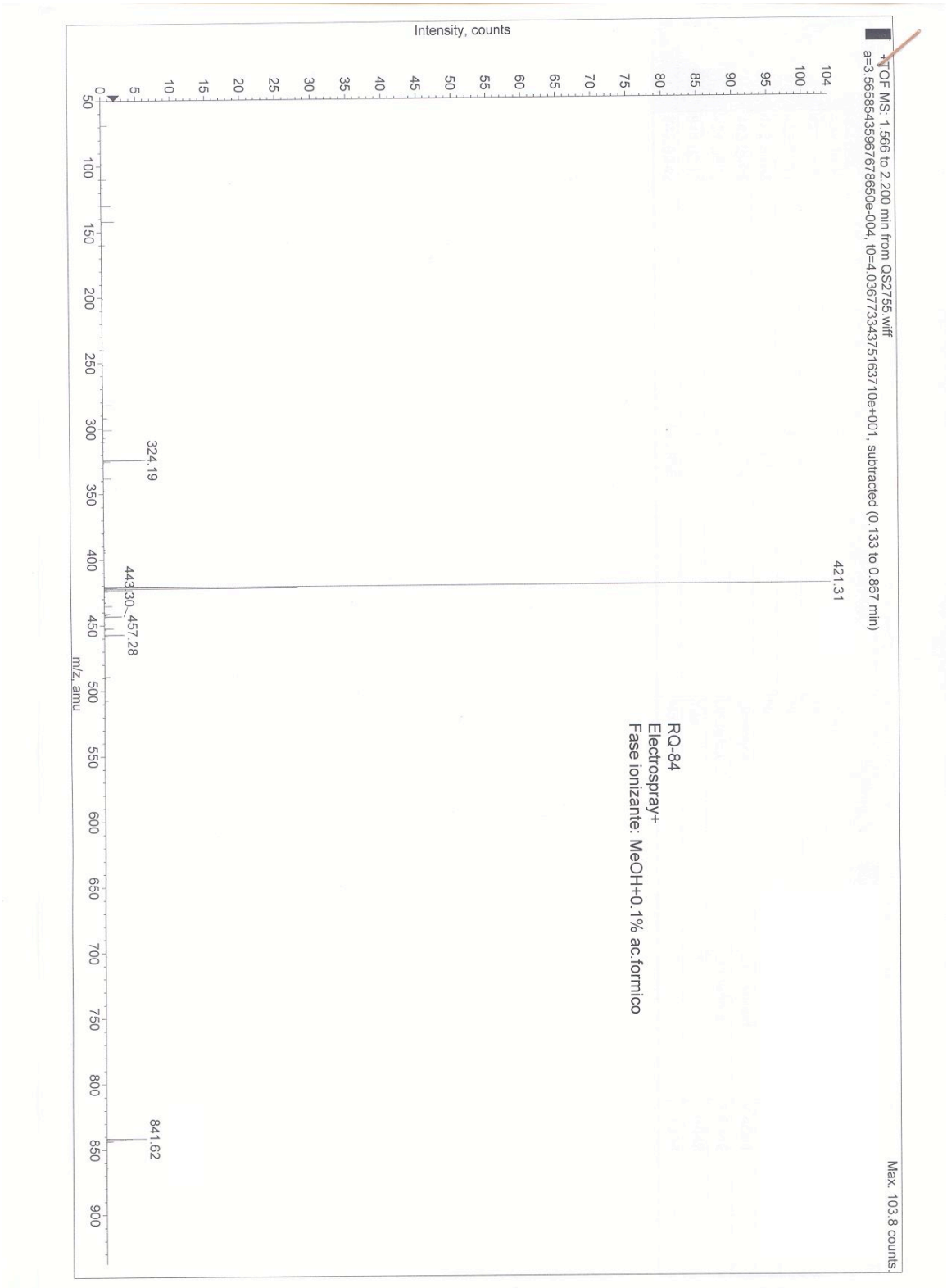
Supplementary figure S5: ^{1}H NMR ($CDCl_3$) spectrum of **4**.



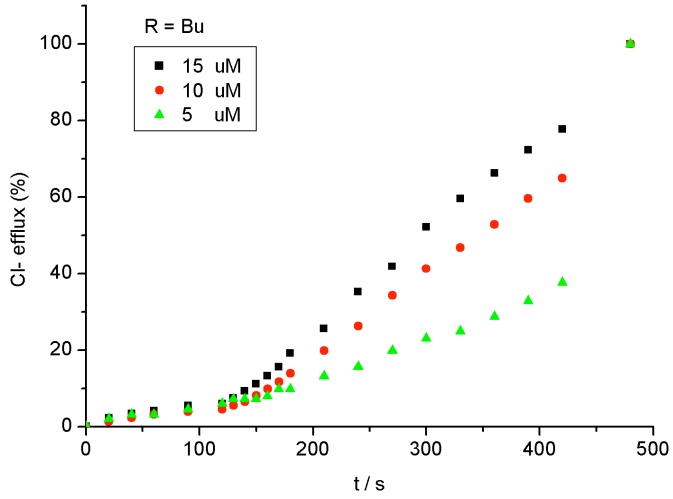
Supplementary figure S6: ^{13}C NMR ($CDCl_3$) spectrum of **4**.



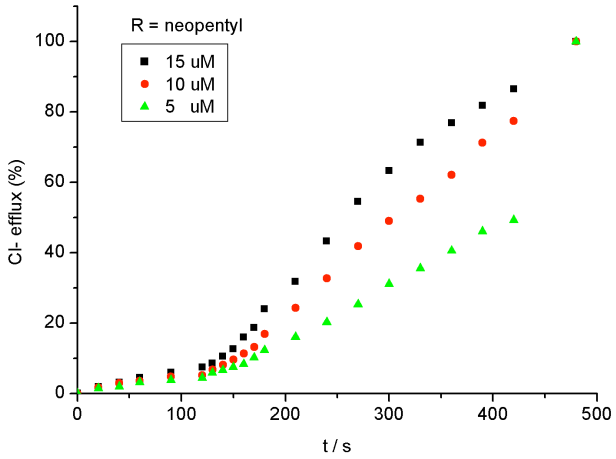
Supplementary figure S7: ^{13}C NMR DEPT ($CDCl_3$) spectrum of **4**.



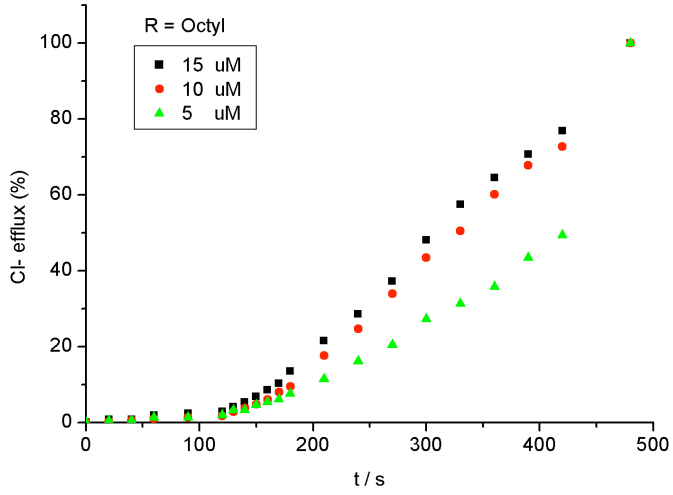
Supplementary figure S8: ESI mass spectrum of **4**.



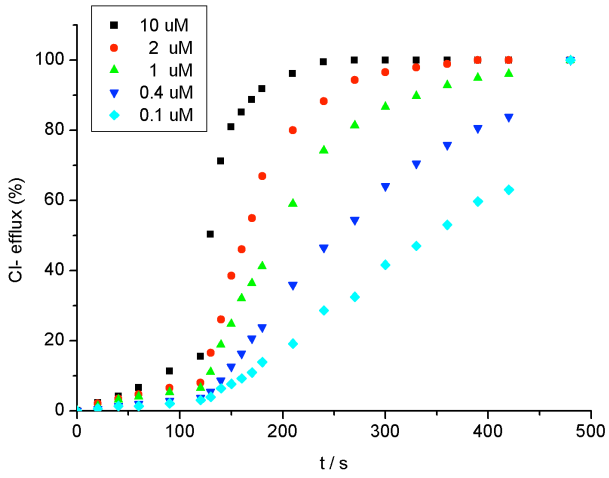
Supplementary figure S9: Chloride efflux promoted upon addition of **2** (15-5μM 1.5-0.5% molar carrier to lipid) to unillamellar POPC vesicles loaded with 451 mM NaCl 20 mM phosphate buffer pH 7.2 dispersed in 150 mM Na_2SO_4 20 mM phosphate buffer pH 7.2. At t = 120 s a solution of $NaHCO_3$ is added to give a 40 mM external concentration. At t = 420 s the vesicles are lysed by addition of detergent and the final reading at t = 540 s considered 100% chloride efflux.



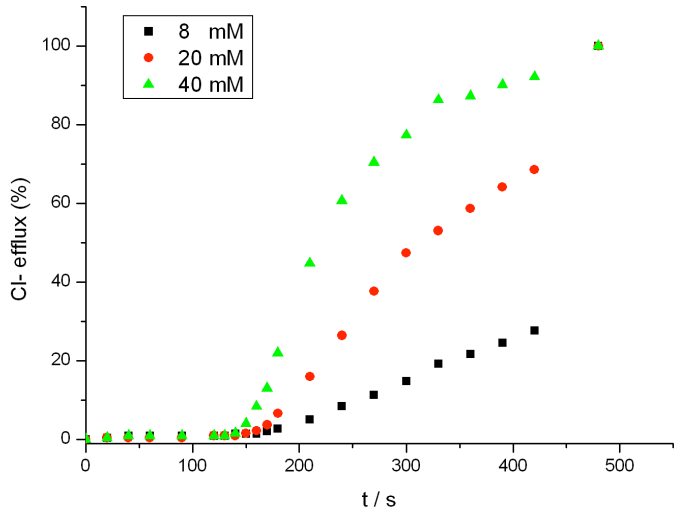
Supplementary figure S10: Chloride efflux promoted upon addition of **3** (15-5μM 1.5-0.5% molar carrier to lipid) to unillamellar POPC vesicles loaded with 451 mM NaCl 20 mM phosphate buffer pH 7.2 dispersed in 150 mM Na_2SO_4 20 mM phosphate buffer pH 7.2. At t = 120 s a solution of $NaHCO_3$ is added to give a 40 mM external concentration. At t = 420 s the vesicles are lysed by addition of detergent and the final reading at t = 540 s considered 100% chloride efflux.



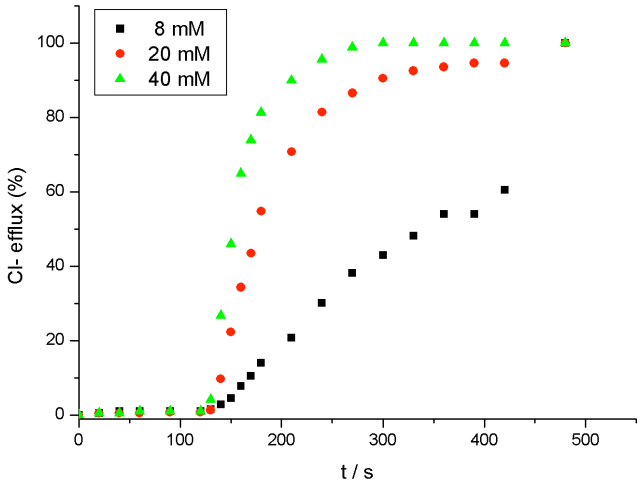
Supplementary figure S11: Chloride efflux promoted upon addition of **4** (15-5μM 1.5-0.5% molar carrier to lipid) to unillamellar POPC vesicles loaded with 451 mM NaCl 20 mM phosphate buffer pH 7.2 dispersed in 150 mM Na_2SO_4 20 mM phosphate buffer pH 7.2. At t = 120 s a solution of $NaHCO_3$ is added to give a 40 mM external concentration. At t = 420 s the vesicles are lysed by addition of detergent and the final reading at t = 540 s considered 100% chloride efflux.



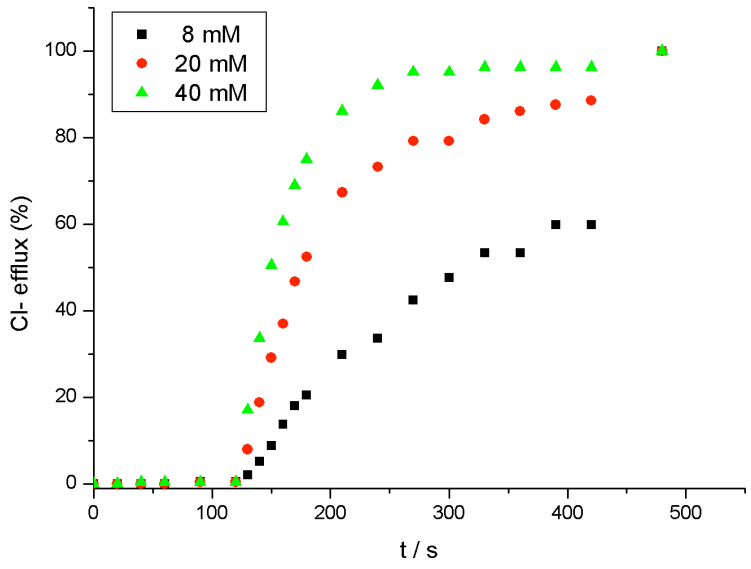
Supplementary figure S12: Chloride efflux promoted upon addition of prodigiosin **1** (10-0.1μM 1-0.01% molar carrier to lipid) to unillamellar POPC vesicles loaded with 451 mM NaCl 20 mM phosphate buffer pH 7.2 dispersed in 150 mM Na_2SO_4 20 mM phosphate buffer pH 7.2. At t = 120 s a solution of $NaHCO_3$ is added to give a 40 mM external concentration. At t = 420 s the vesicles are lysed by addition of detergent and the final reading at t = 540 s considered 100% chloride efflux.



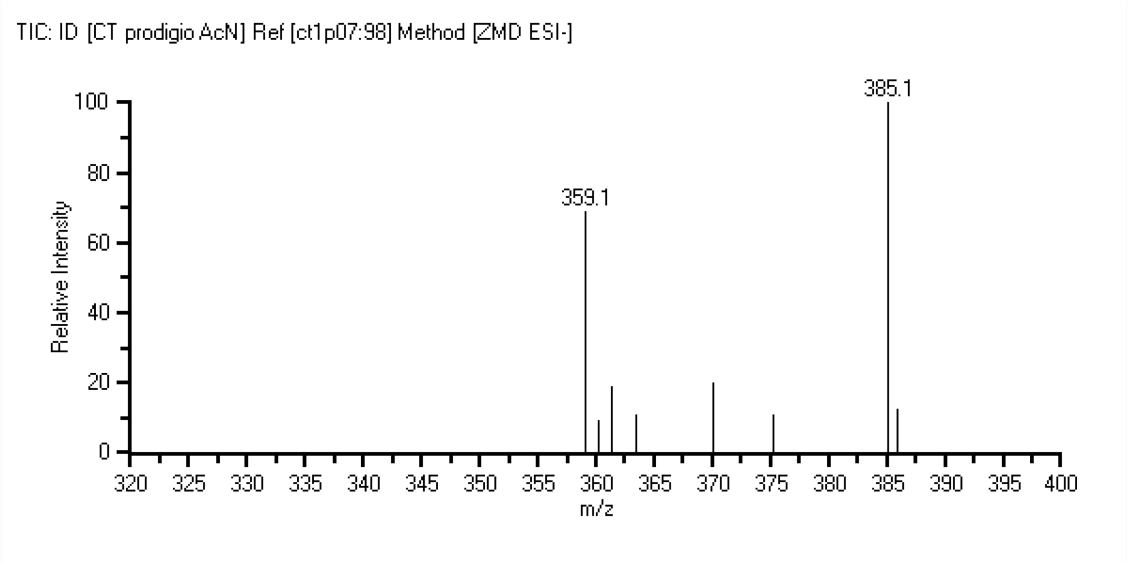
Supplementary figure S13: Chloride efflux upon addition of of 4,6-dihydroxy-*N,N'*-butyl isophthalamide **2** to vesicles composed of POPC. The vesicles contained NaCl (488 mM) and were immersed in Na_2SO_4 (166 mM), pH 7.0 solution; at 120 s, a $NaNO_3$ solution to give a nitrate concentration of 8-40mM was added and at 420 s the vesicles were lysed to obtain 100% chloride efflux.



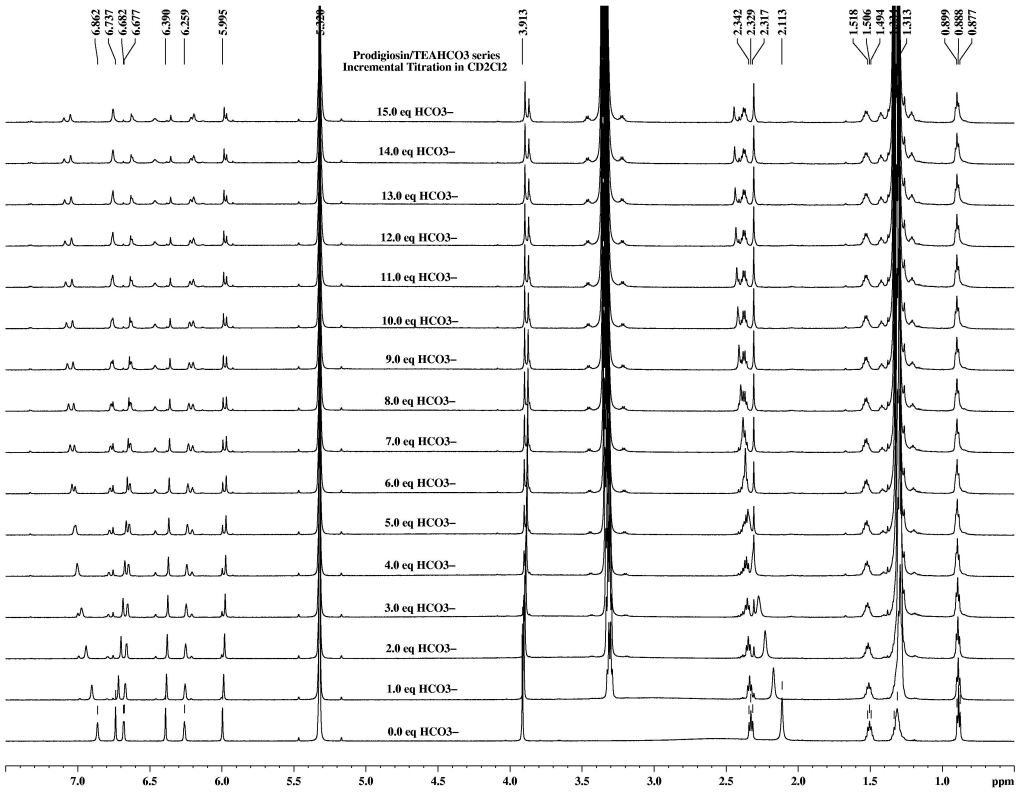
Supplementary figure S14: Chloride efflux upon addition of of 4,6-dihydroxy-*N,N'*-neopentyl isophthalamide **3** to vesicles composed of POPC. The vesicles contained NaCl (500 mM) and were immersed in Na_2SO_4 (333 mM), pH 7.0 solution; at 120 s, a $NaNO_3$ solution to give a nitrate concentration of 8-40mM was added and at 420 s the vesicles were lysed to obtain 100% chloride efflux.



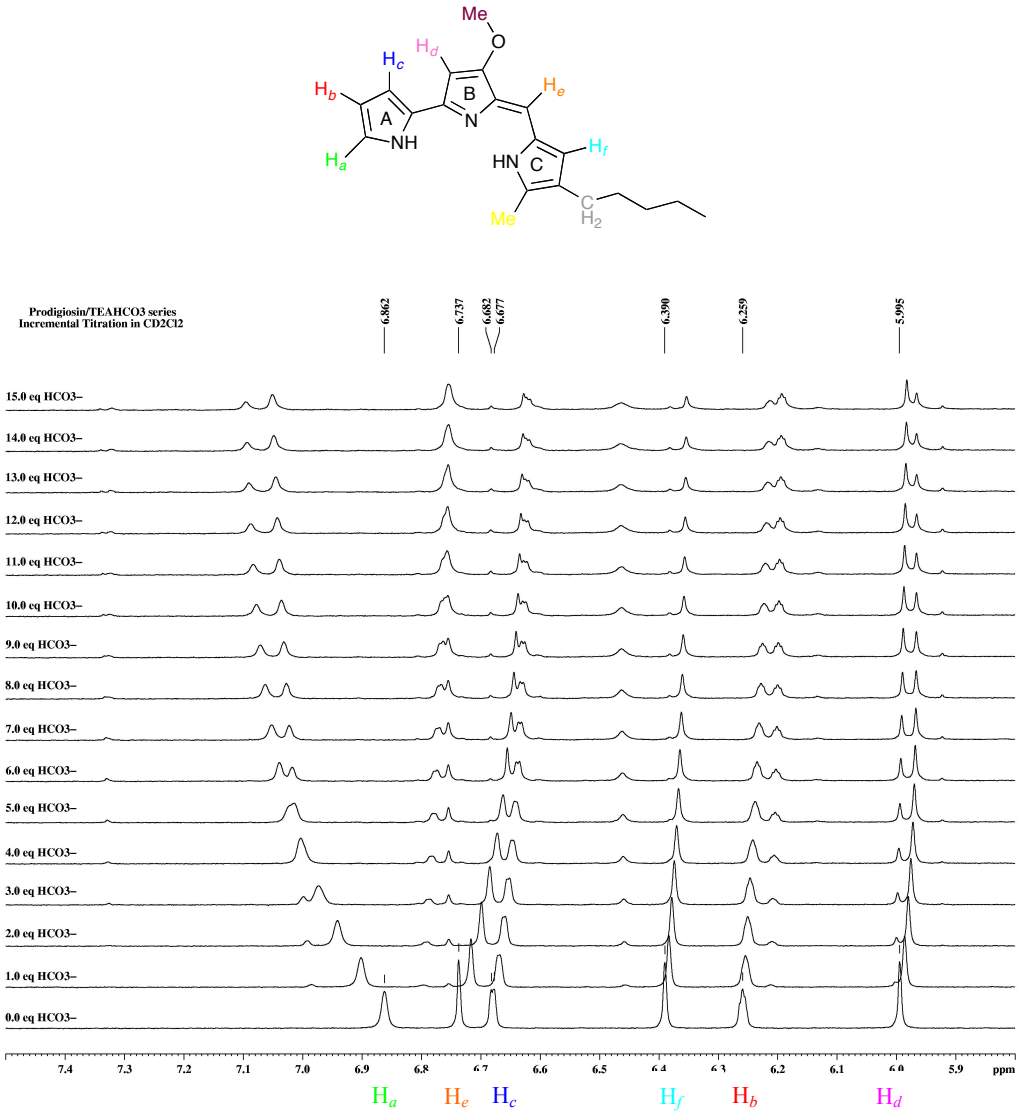
Supplementary figure S15: Chloride efflux upon addition of of 4,6-dihydroxy-*N,N'*-octyl isophthalamide **4** to vesicles composed of POPC. The vesicles contained NaCl (500 mM) and were immersed in Na_2SO_4 (333 mM), pH 7.0 solution; at 120 s, a $NaNO_3$ solution to give a nitrate concentration of 8-40mM was added and at 420 s the vesicles were lysed to obtain 100% chloride efflux.



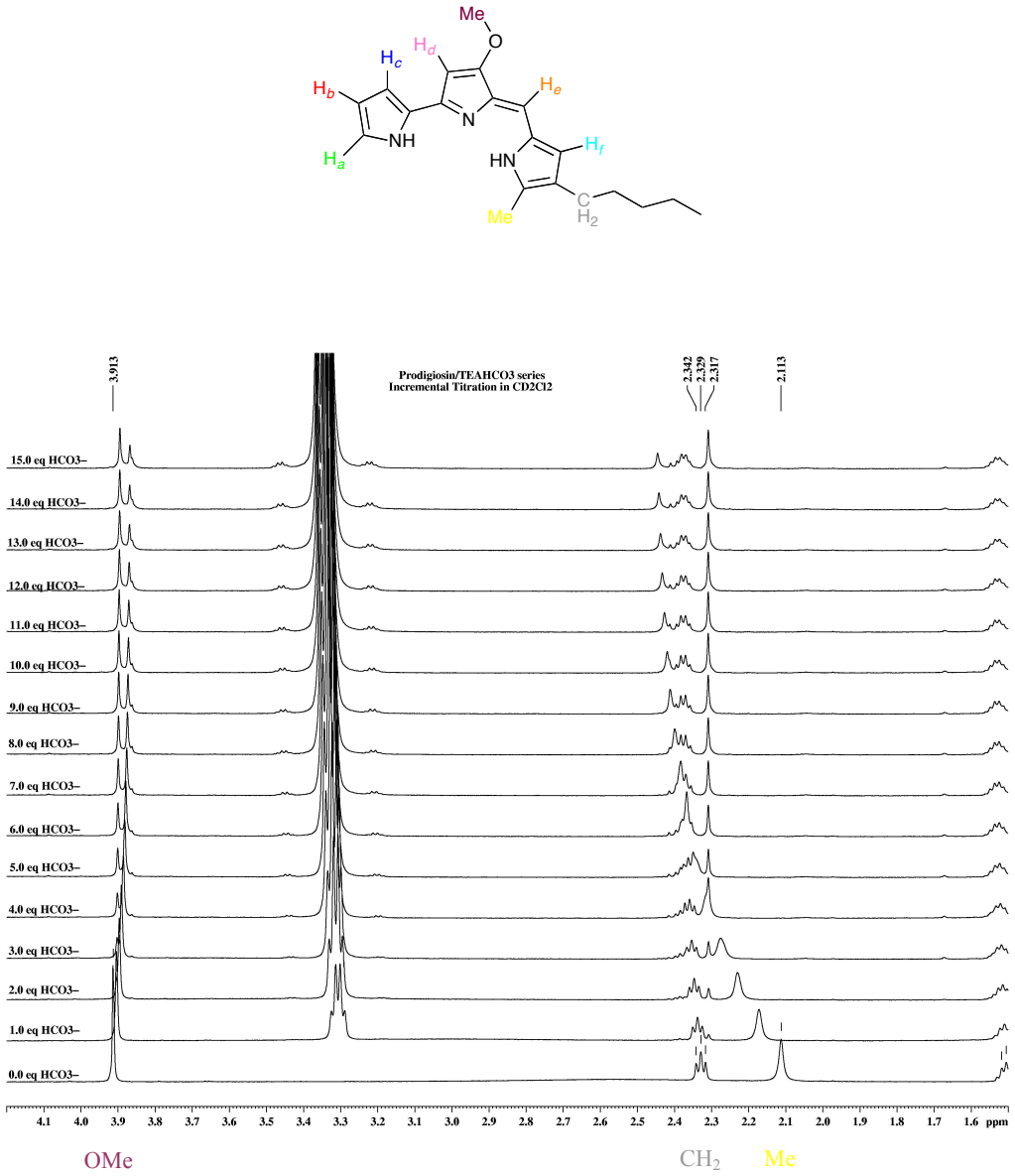
Supplementary figure S16: Electrospray mass spectrometry (negative mode) of prodigiosin **1** showing chloride and bicarbonate complexes.



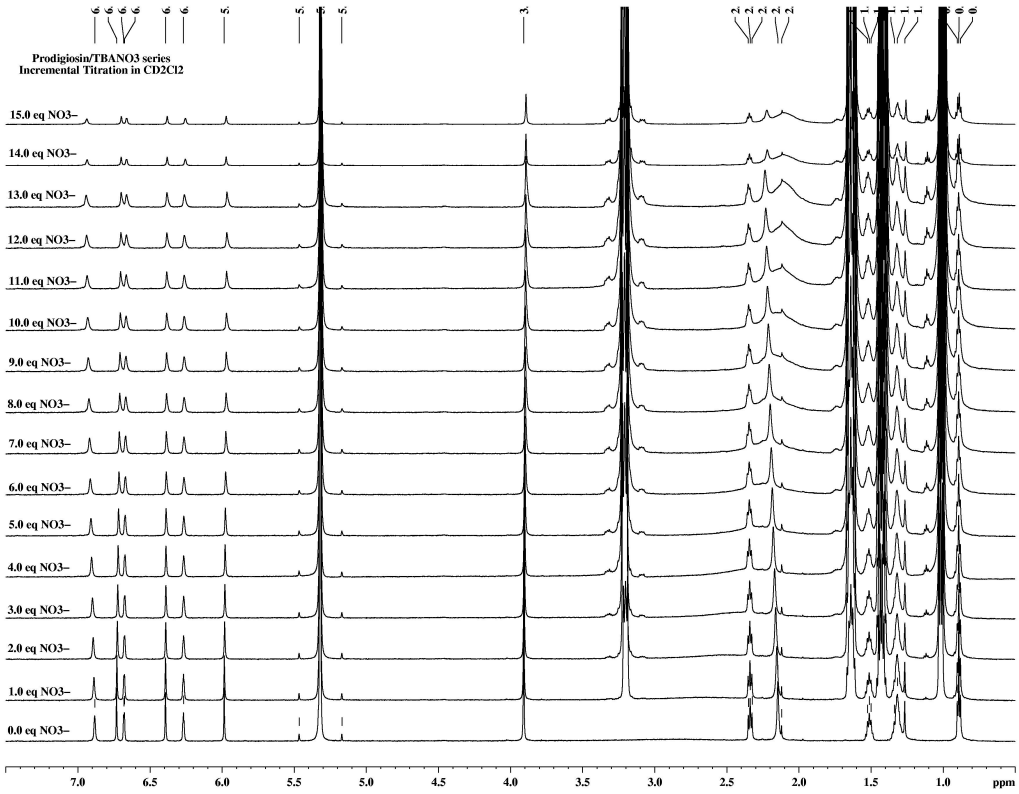
Supplementary figure S17: ^{1}H NMR titration of prodigiosin **1** with tetraethylammonium bicarbonate in CD_2Cl_2.



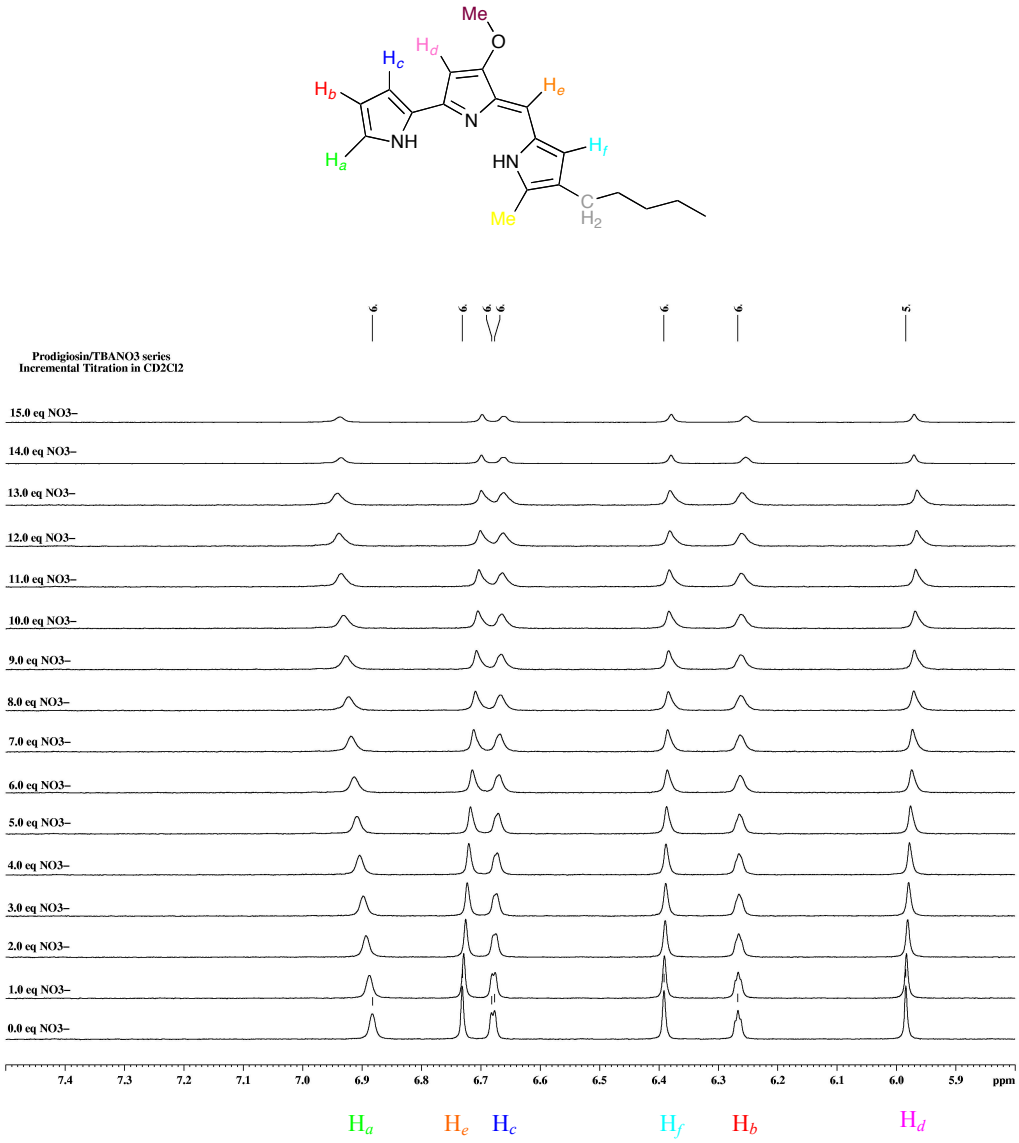
Supplementary figure S18: Aromatic region of the ^{1}H NMR titration of prodigiosin **1** with tetraethylammonium bicarbonate



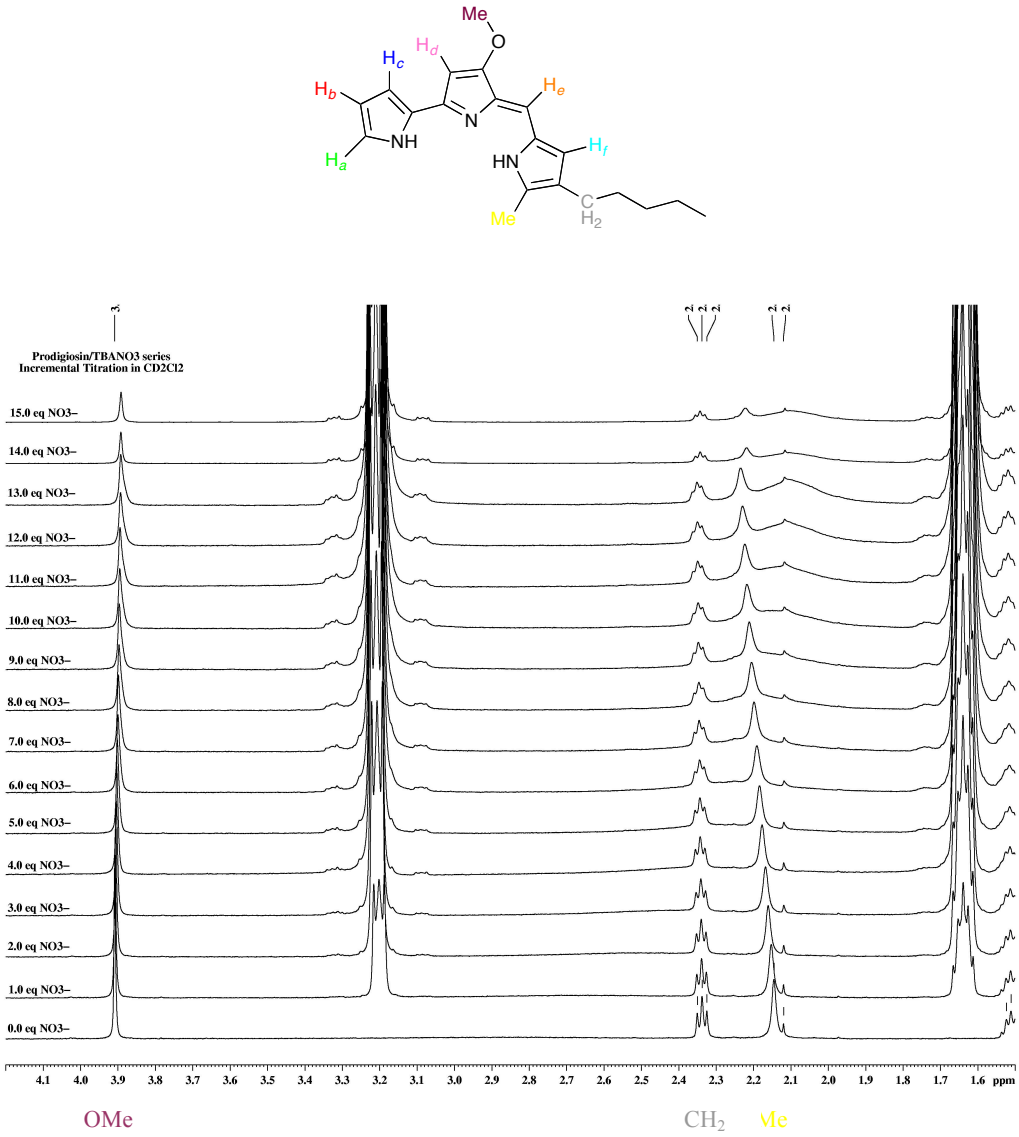
Supplementary figure S19: Aliphatic region of the ^{1}H NMR titration of prodigiosin **1** with tetraethylammonium bicarbonate



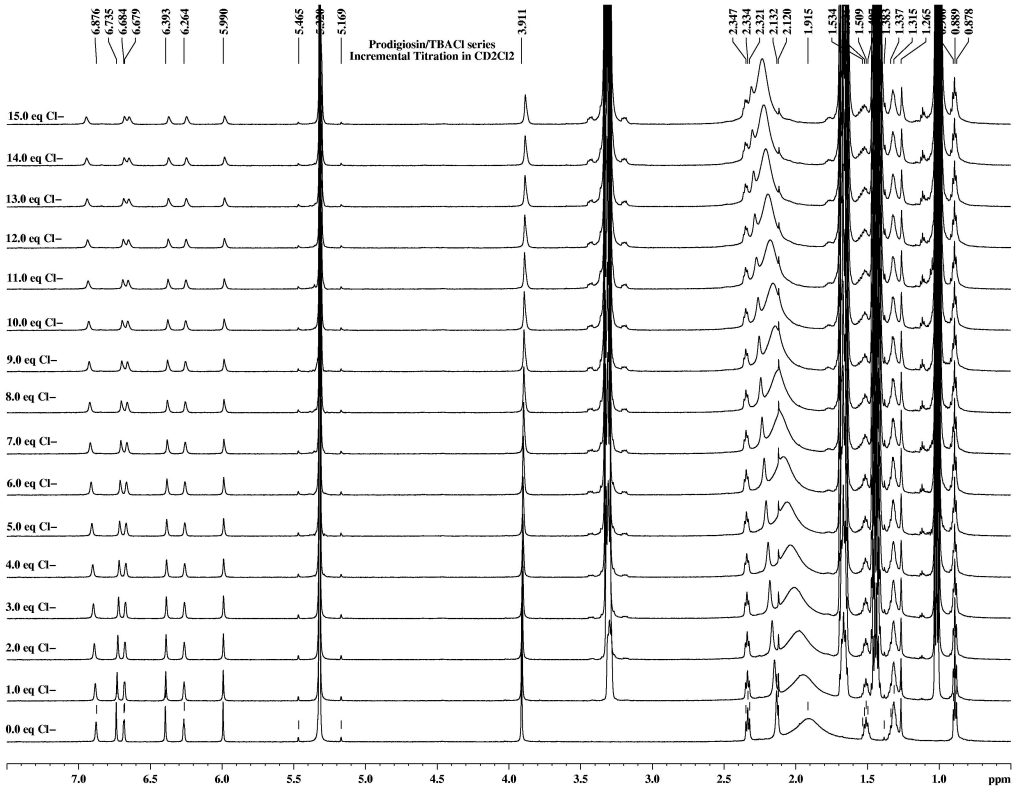
Supplementary figure S20: ^{1}H NMR titration of prodigiosin **1** with tetrabutylammonium nitrate in CD_2Cl_2.



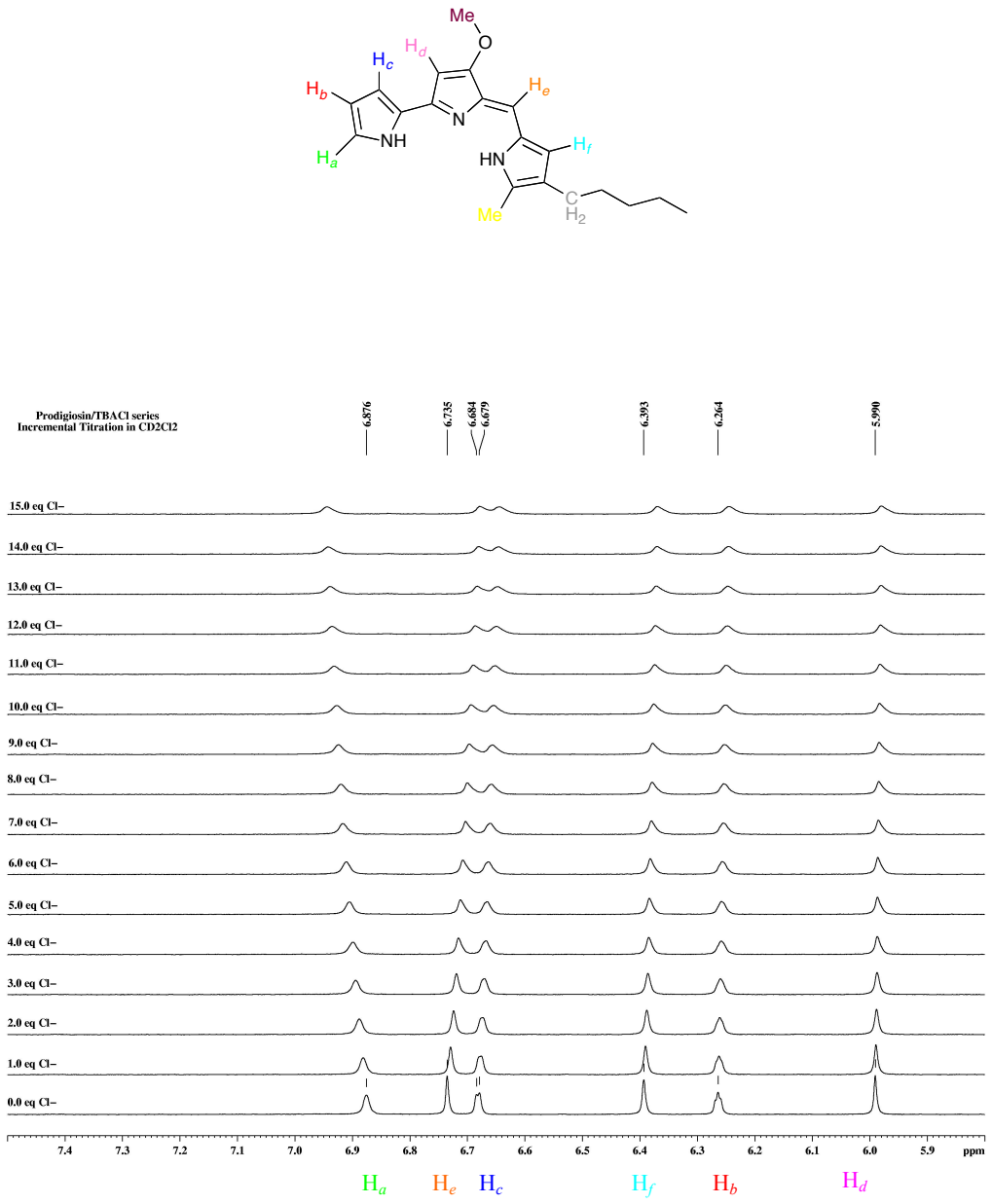
Supplementary figure S21: Aromatic region of the ^{1}H NMR titration of prodigiosin **1** with tetrabutylammonium nitrate in CD_2Cl_2.



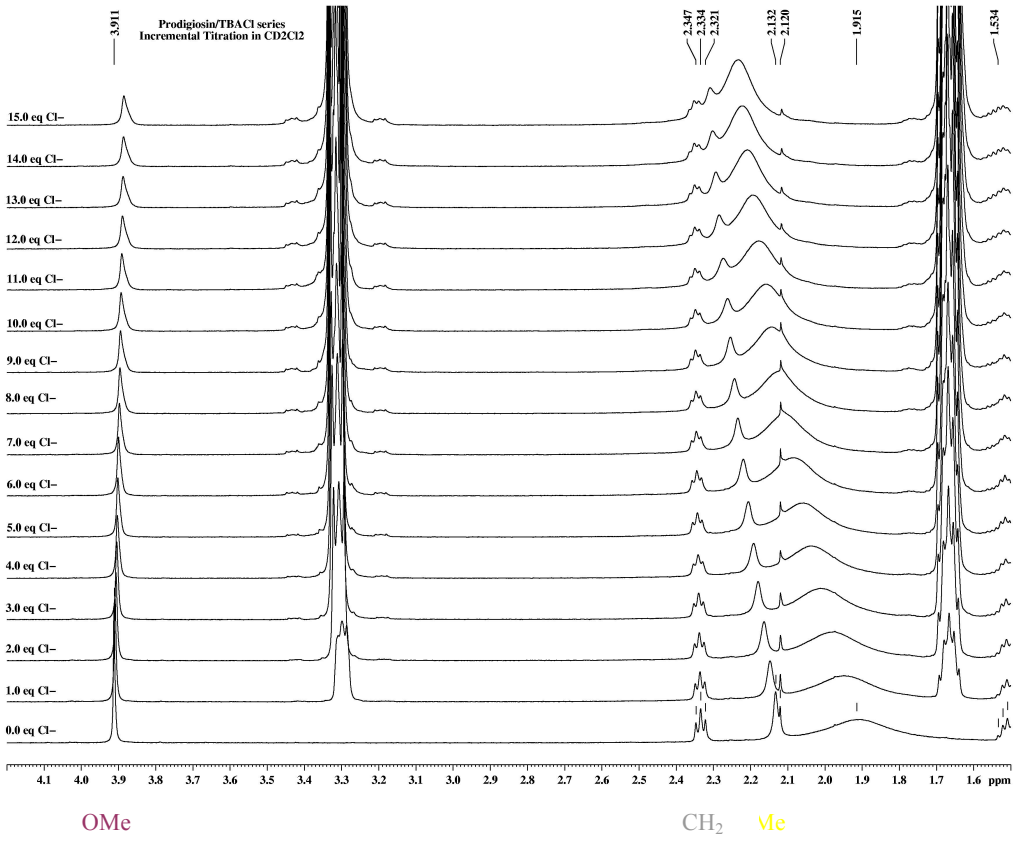
Supplementary figure S22: Aliphatic region of the ^{1}H NMR titration of prodigiosin **1** with tetrabutylammonium nitrate in CD_2Cl_2.



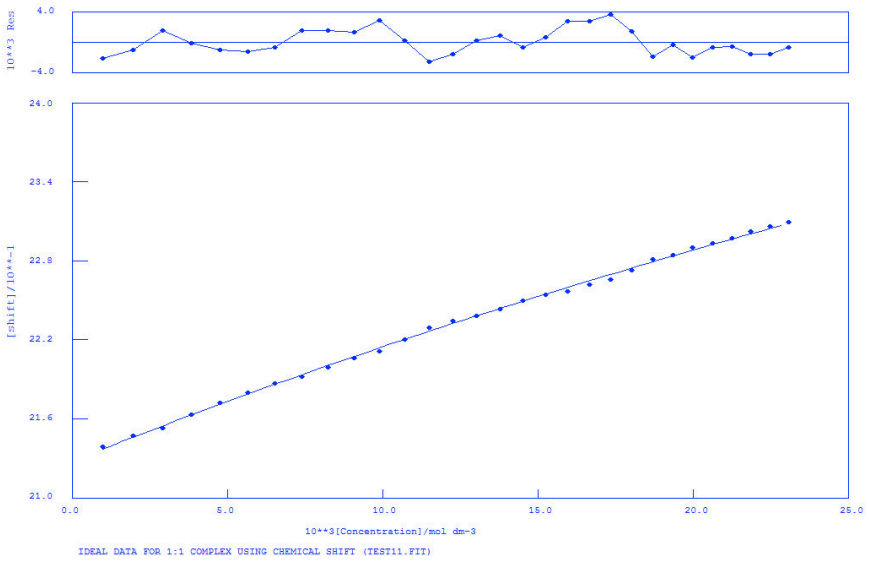
Supplementary figure S23: ^{1}H NMR titration of prodigiosin **1** with tetrabutylammonium chloride in CD_2Cl_2.



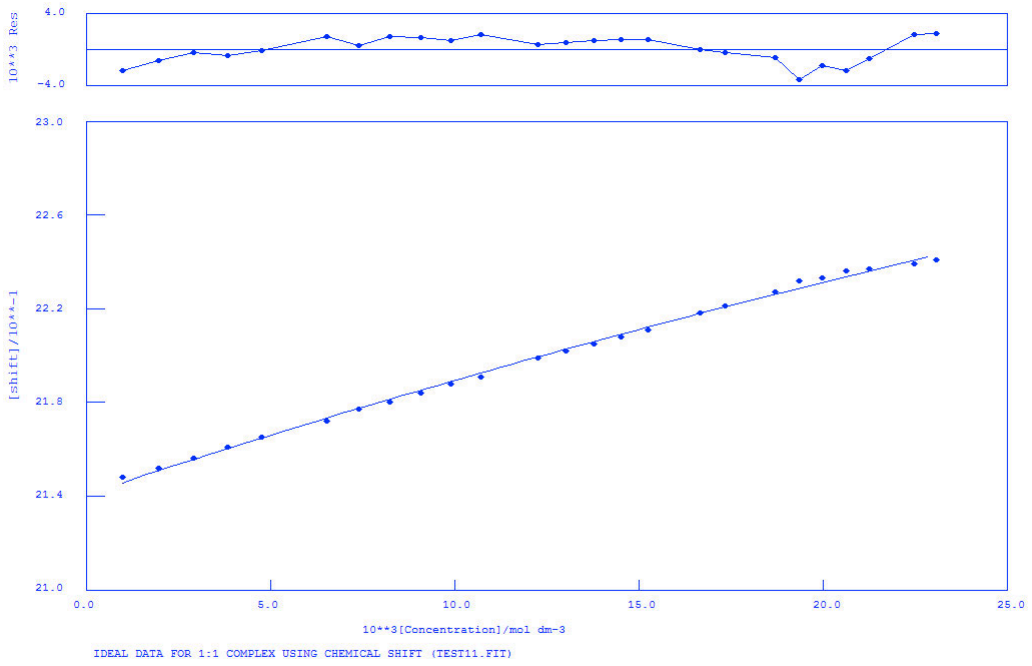
Supplementary figure S24: Aromatic region of the ^{1}H NMR titration of prodigiosin **1** with tetrabutylammonium chloride in CD_2Cl_2.



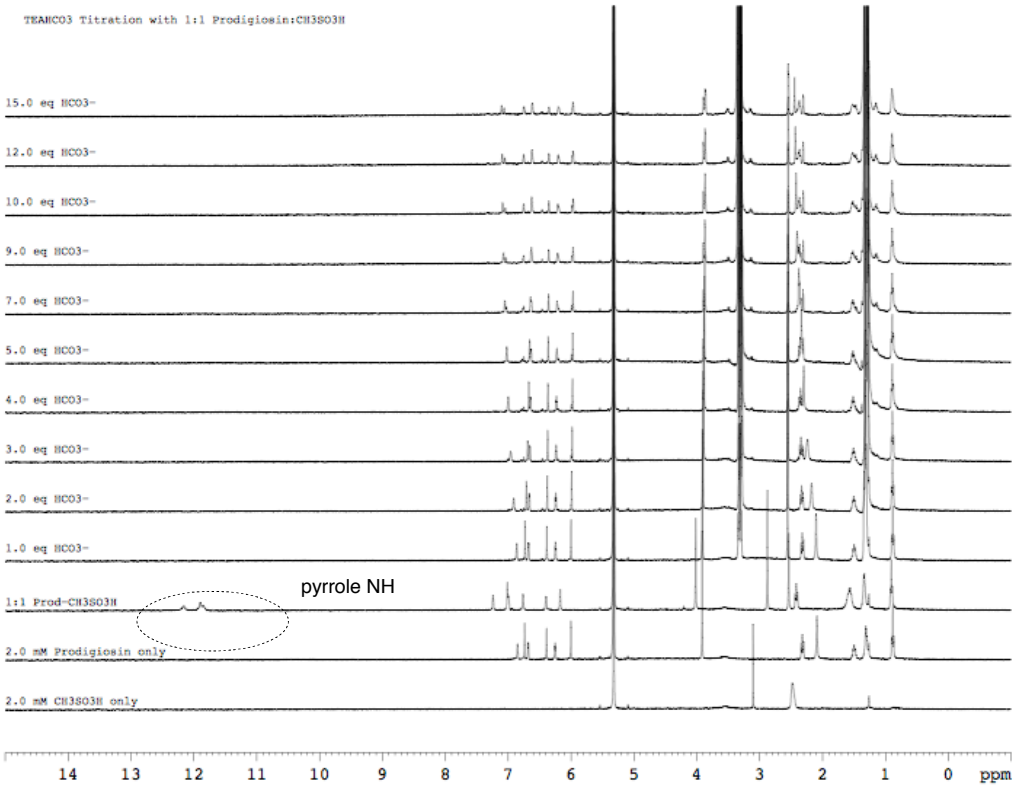
Supplementary figure S25: Aliphatic region of the ^{1}H NMR titration of prodigiosin **1** with tetrabutylammonium chloride in CD_2Cl_2.



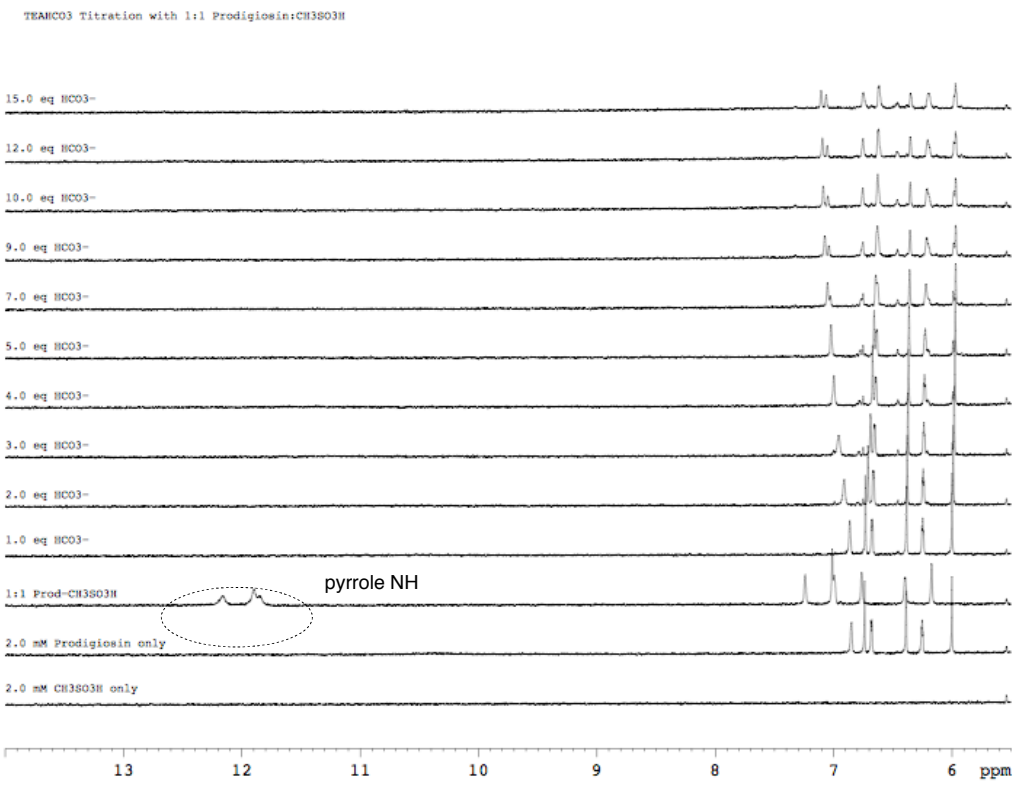
Supplementary figure S26: ^{1}H NMR titration of prodigiosin **1** with tetrabutylammonium chloride in CD_2Cl_2. K_a = 9.7 ± 1.4 M^{-1}



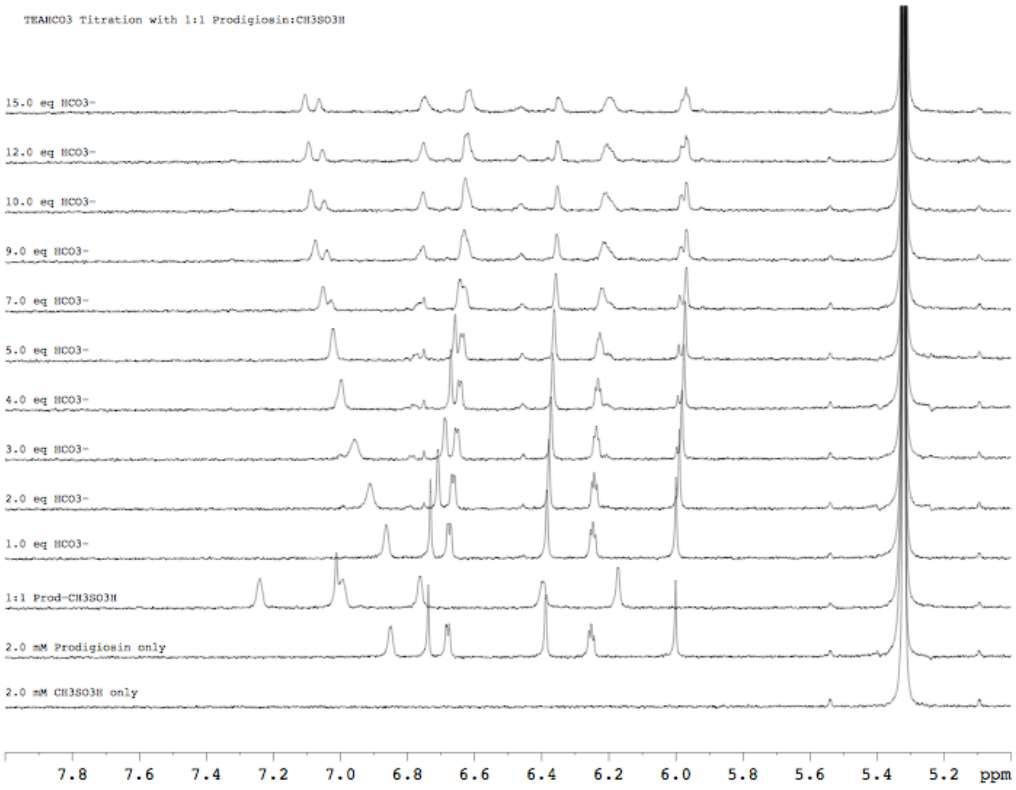
Supplementary figure S27: ^{1}H NMR titration of prodigiosin **1** with tetrabutylammonium nitrate in CD_2Cl_2. K_a = 8.9 ± 1.9 M^{-1}



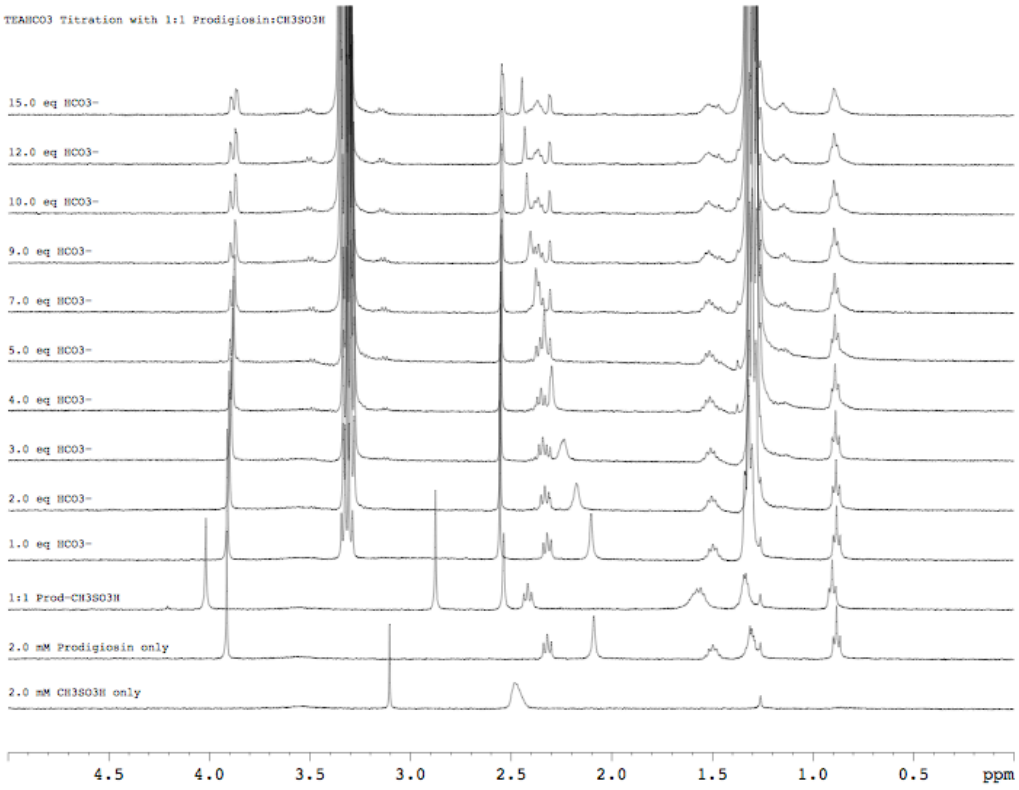
Supplementary figure S28: ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetraethylammonium bicarbonate in CD_2Cl_2.



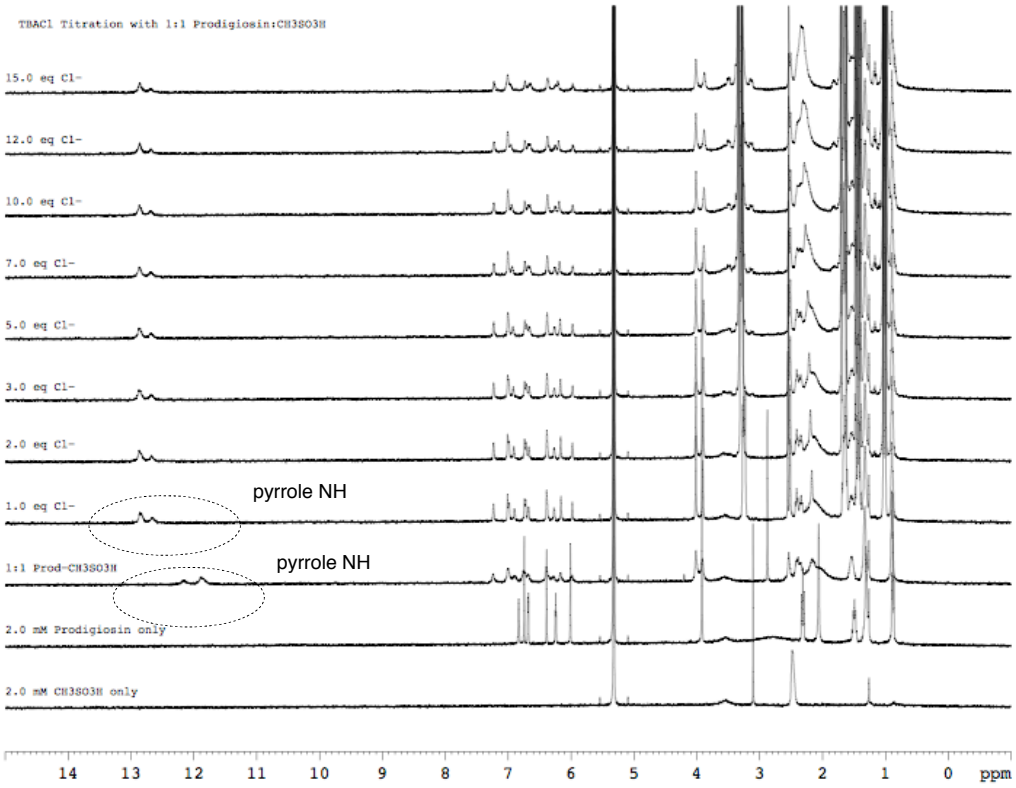
Supplementary figure S29: Aromatic and pyrrole NH region in the ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetraethylammonium bicarbonate in CD_2Cl_2.



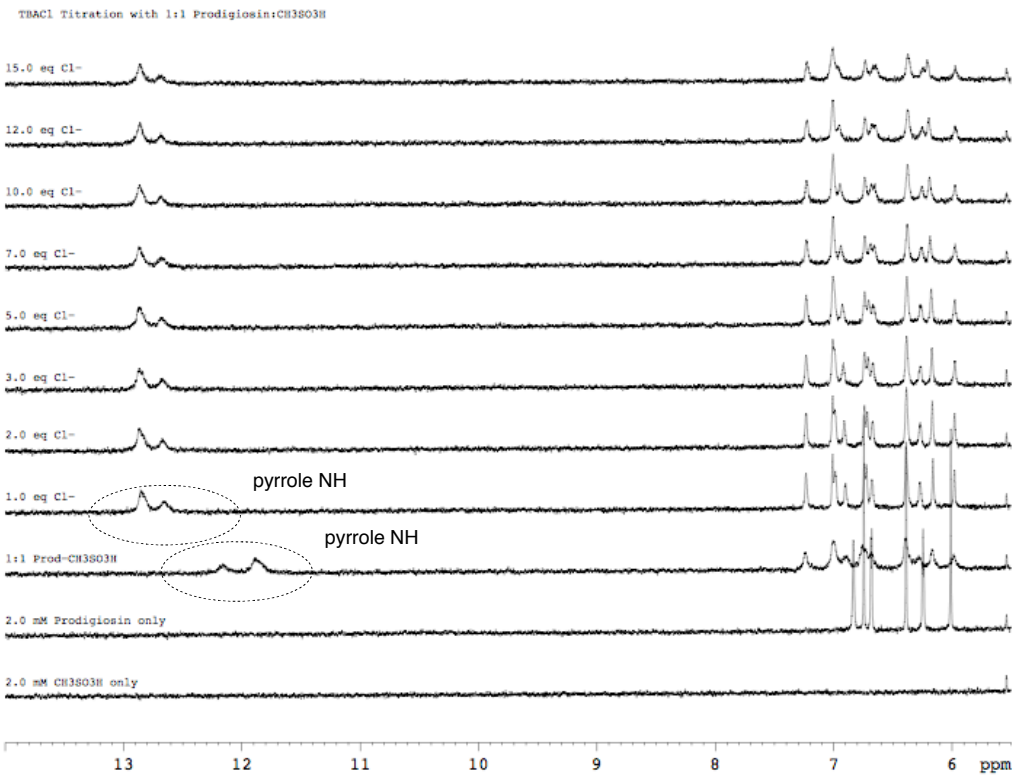
Supplementary figure S30: Aromatic region in the ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetraethylammonium bicarbonate in CD_2Cl_2.



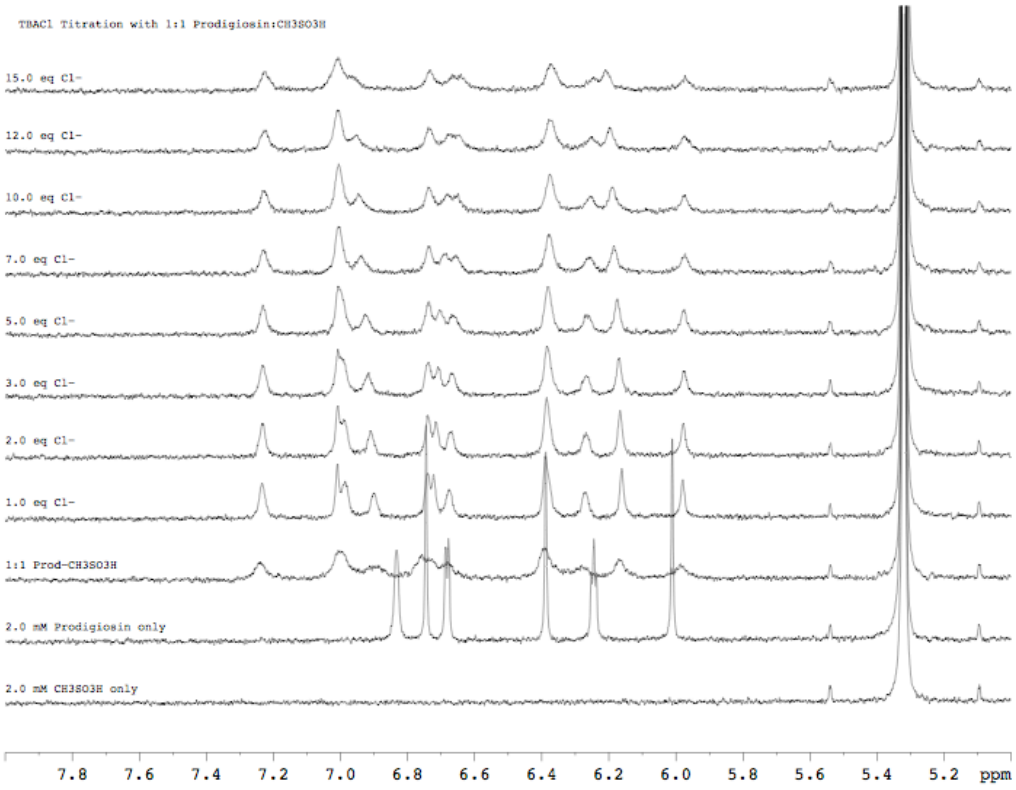
Supplementary figure S31: Aliphatic region in the ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetraethylammonium bicarbonate in CD_2Cl_2.



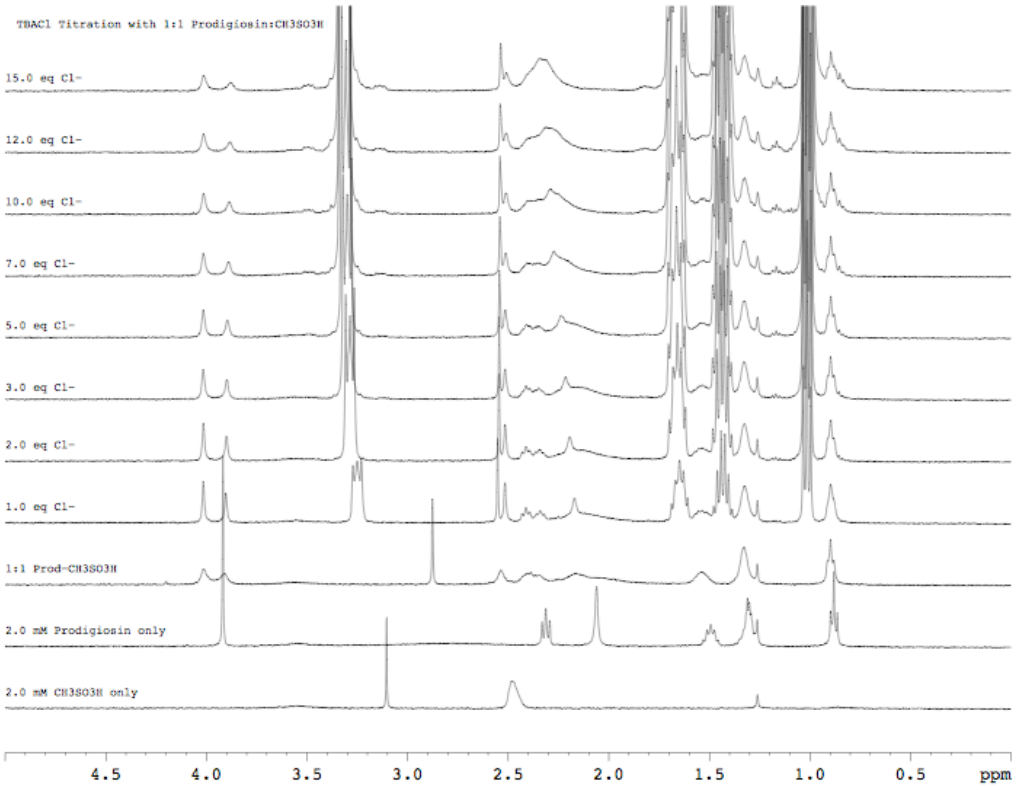
Supplementary figure S32: ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetrabutylammonium chloride in CD_2Cl_2.



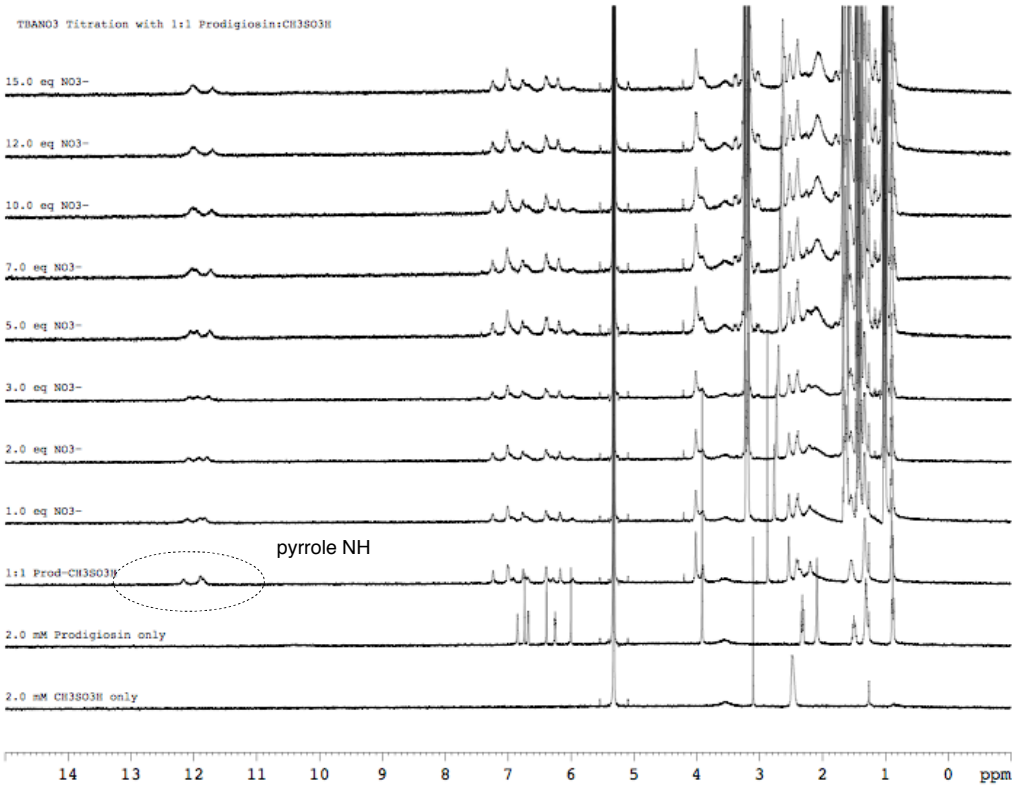
Supplementary figure S33: Aromatic and pyrrole NH region of the ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetrabutylammonium chloride in CD_2Cl_2.



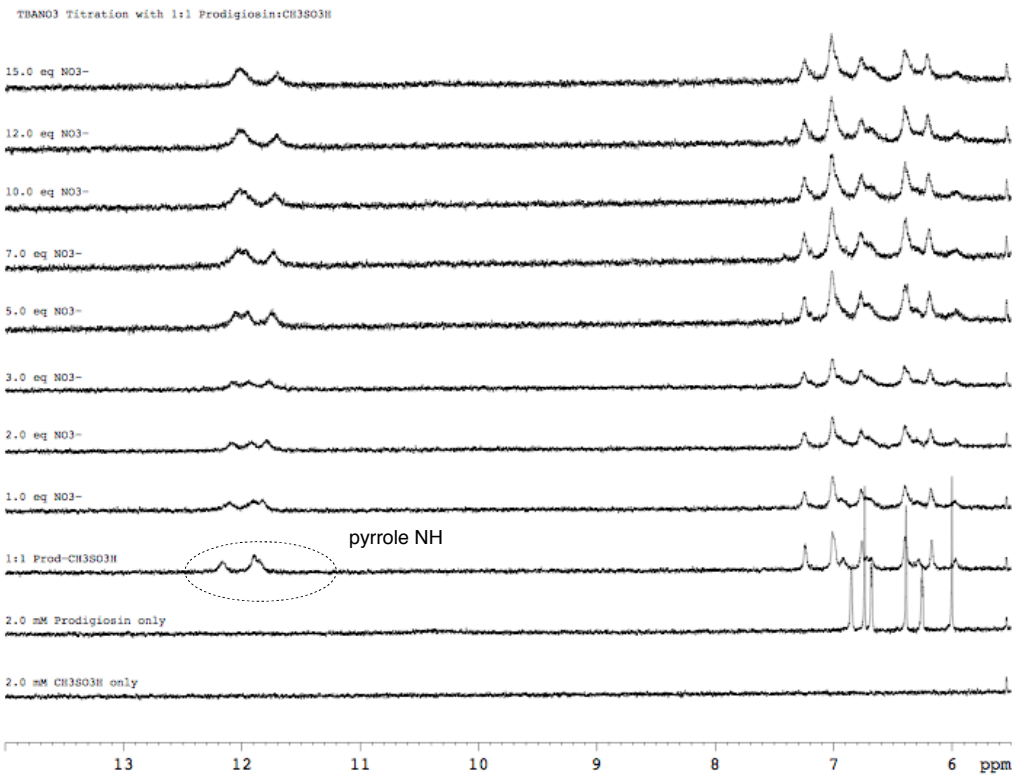
Supplementary figure S34: Aromatic region of the ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetrabutylammonium chloride in CD_2Cl_2.



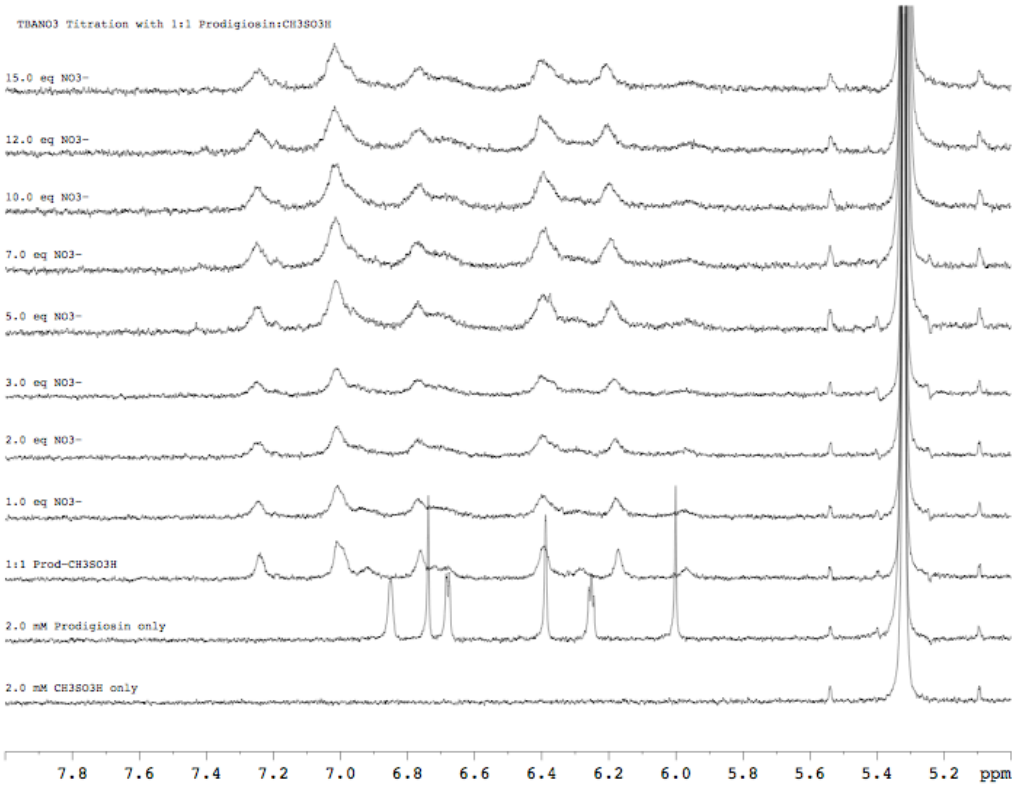
Supplementary figure S35: Aliphatic region of the ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetrabutylammonium chloride in CD_2Cl_2.



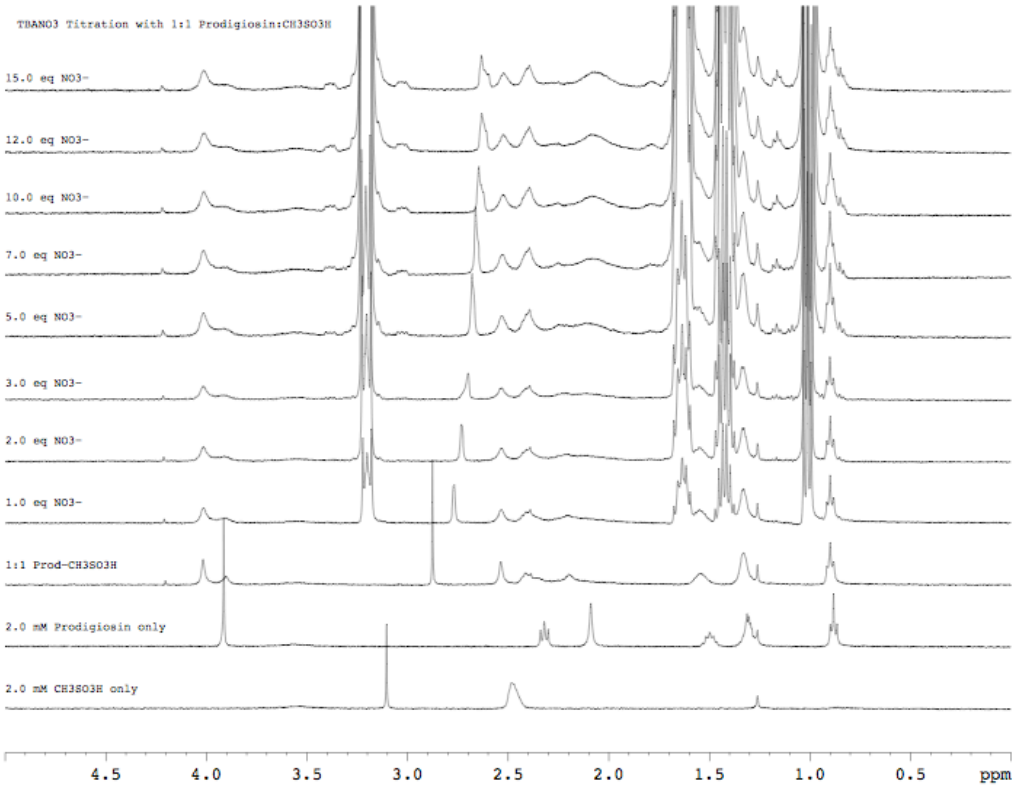
Supplementary figure S36: ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetrabutylammonium nitrate in CD_2Cl_2.



Supplementary figure S37: Aromatic and pyrrole NH region of the ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetrabutylammonium nitrate in CD_2Cl_2.



Supplementary figure S38: Aromatic region of the ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetrabutylammonium nitrate in CD_2Cl_2.



Supplementary figure S39: Aliphatic region of the ^{1}H NMR titration of prodigiosin **1** + CH_3SO_3H with tetrabutylammonium nitrate in CD_2Cl_2.

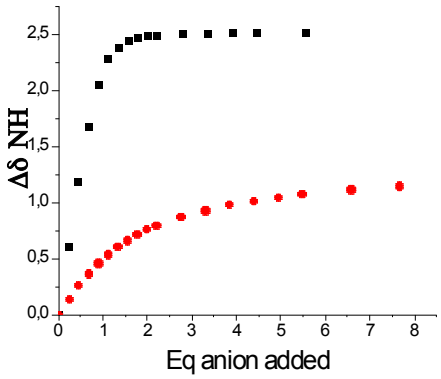
NMR studies with isophthalamides

We first investigated the anion binding properties of N,N`butyl-4,6-dihydroxyisophthalamide **2** using NMR titration techniques (Figure S40). In acetonitrile solution the receptor shows good selectivity for chloride against nitrate (Table S1). Using more competitive solvents such as DMSO and DMSO with increasing amounts of water, the stability constants are lower due to the more competitive solvent mixture (Figure S41). An NMR titration experiment in DMSO with tetraethylammonium bicarbonate showed that hydroxyl groups deprotonate under these conditions so no stability constant under these conditions could be obtained.

Table S1. Stability constants K_a (M^{-1}) of receptor **2** toward chloride[a] calculated by NMR titration experiments at 25 ºC.

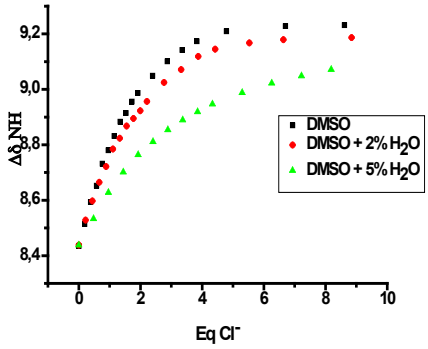
Receptor	Cl^- (CD_3CN)	Cl^- (d_6-DMSO)	NO_3^- (CD_3CN)	NO_3^- (d_6-DMSO)
K	5231.48	70.0	94.94	<10
error	478.7	3.15	1.725	-

[a]Anion added as tetrabutylammonium salt.



Supplementary Figure S40. Chemical shifts induced in the N-H group by the addition of increasing amounts of TBACl (black dots) and $TBANO_3$ (red dots) in CD_3CN

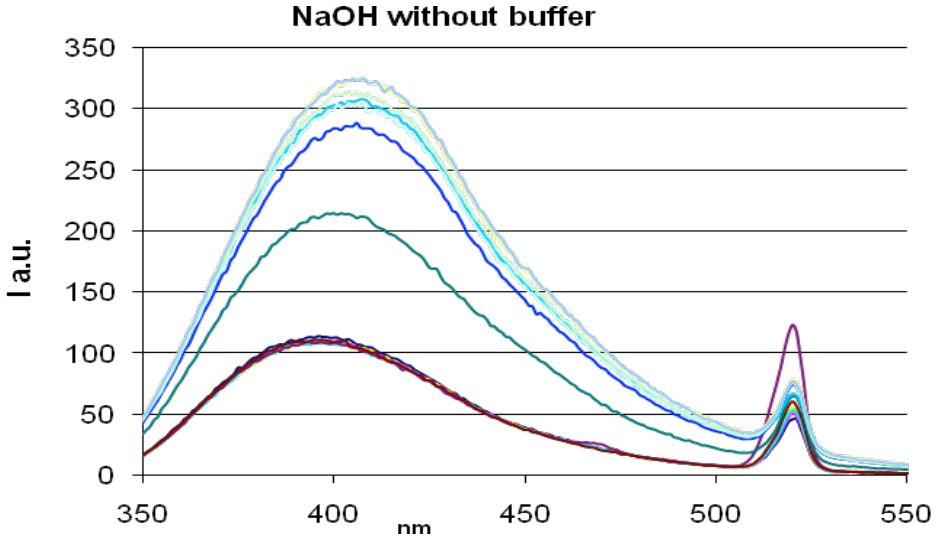
	$\Delta\delta_{NH}$	K_a (M^{-1}) [a]
d_6-DMSO	0.79	70
d_6-DMSO + 2% H_2O	0.74	64
d_6-DMSO + 5% H_2O	0.66	46
a) Error < 10 %.		



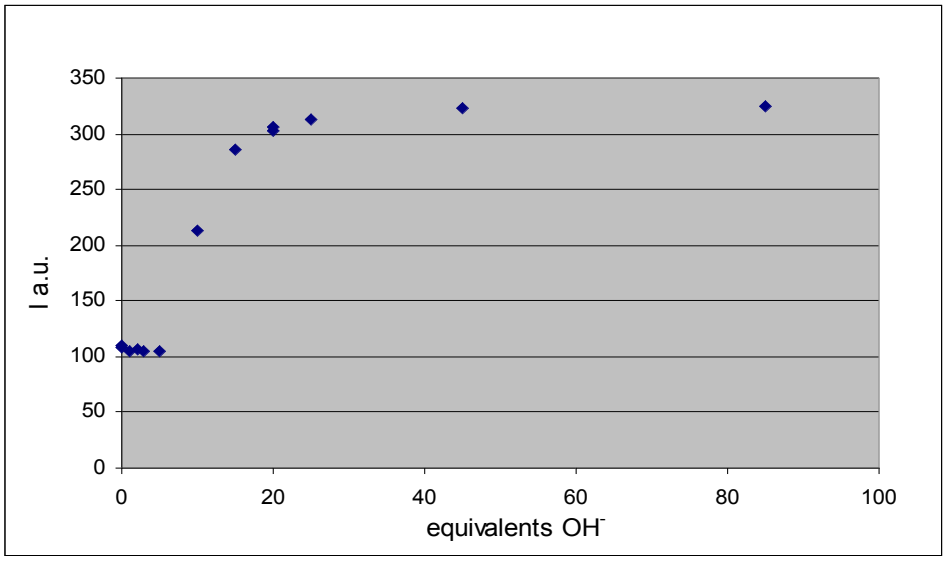
Supplementary Figure S41. Chemical shifts induced in the N-H group of receptor **2** by the addition of increasing amounts of TBACl in d_6-DMSO with increasing amounts of water.

Fluorescence experiments.

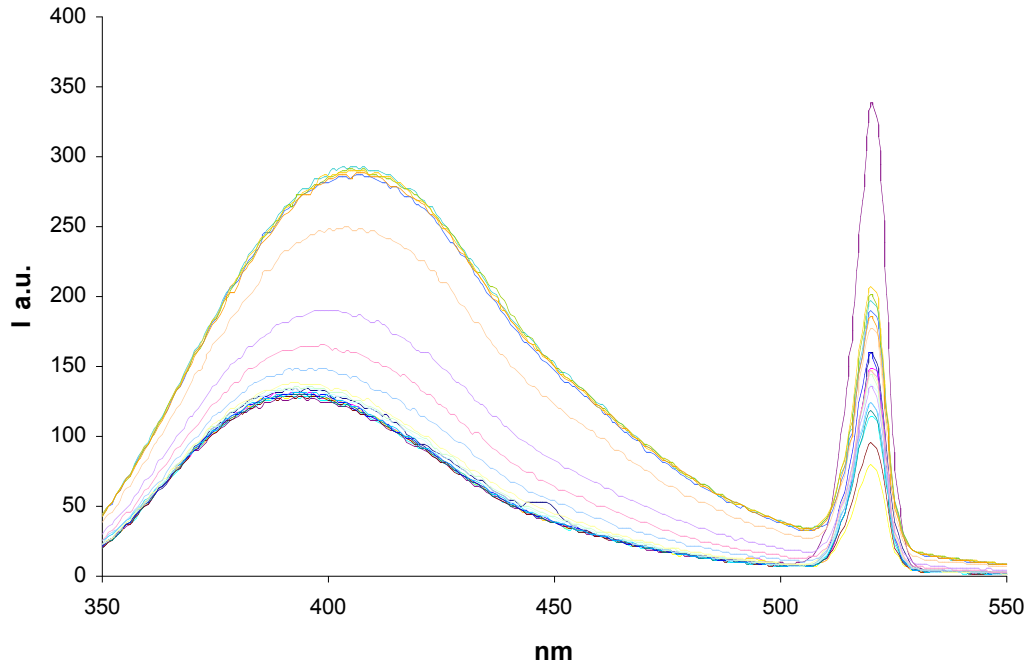
We decided to explore the interaction of compound **2** with different anions using fluorescence techniques. A solution of N,N´dibutyl-4,6-dihydroxyisophthalamide **2** (10^{-5} M) in a mixture water/DMSO 98:2 v :v (HEPES buffer 5mM pH=7.2) was excited using λ_{ex} = 260nm, and the fluorescence response monitored upon addition of increasing amounts of anions added as sodium salts in water. Deprotonation of the hydroxyl groups is easily identified as a dramatic increase of the fluorescence emission results from the rigidification of the molecule (due to $NH^{\cdots}O^{-}$ hydrogen bond formation), as a titration experiment with NaOH in the absence of buffer shows (Figure S42, S43). When the experiments were repeated in the presence of buffer, addition of NaOH resulted in no changes to the point of the buffer exhaustion, followed by deprotonation (Figure S44, S45). In a buffered solution, addition of $NaHCO_3$ to concentrations of receptor similar to those used in the transport experiments resulted in no deprotonation, and only minor changes in the fluorescence emission occurred (Figure S46, S47). The same result is obtained by adding NaCl (Figure S48, S49). These results demonstrate that isophthalamide **2** does not deprotonate in the presence of bicarbonate under the conditions of the membrane transport experiments.



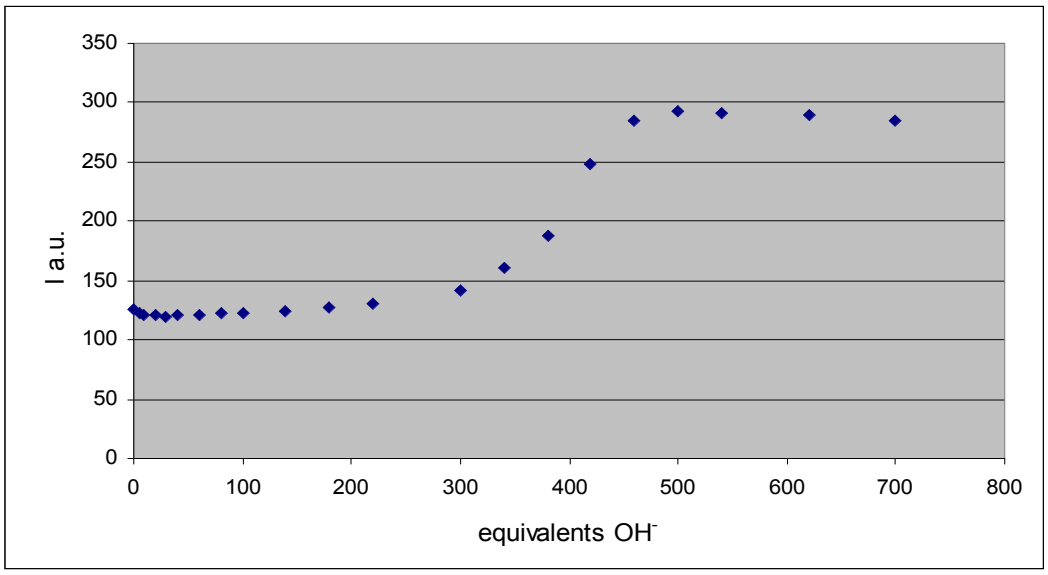
Supplementary Figure S42. Changes observed in the fluorescence spectra of **2** (10^{-5} M water DMSO 98/2 v/v) upon addition of NaOH in water. (λ_{ex} = 260nm)



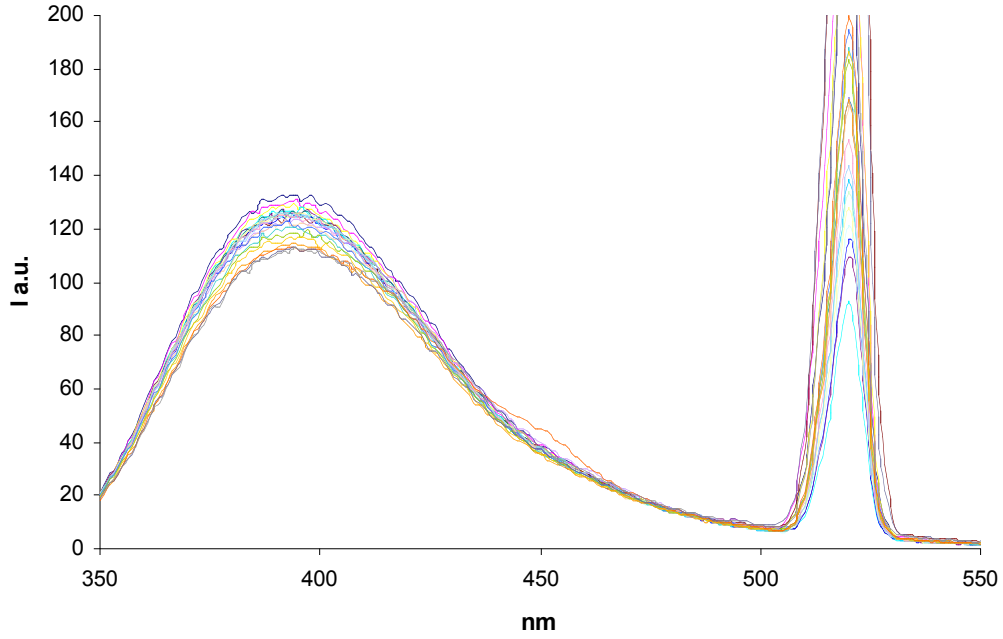
Supplementary Figure S43. Titration profile of changes observed in the fluorescence spectra of **2** (10^{-5} M water DMSO 98/2 v/v) upon addition of NaOH in water. (λ_{ex} = 260nm)



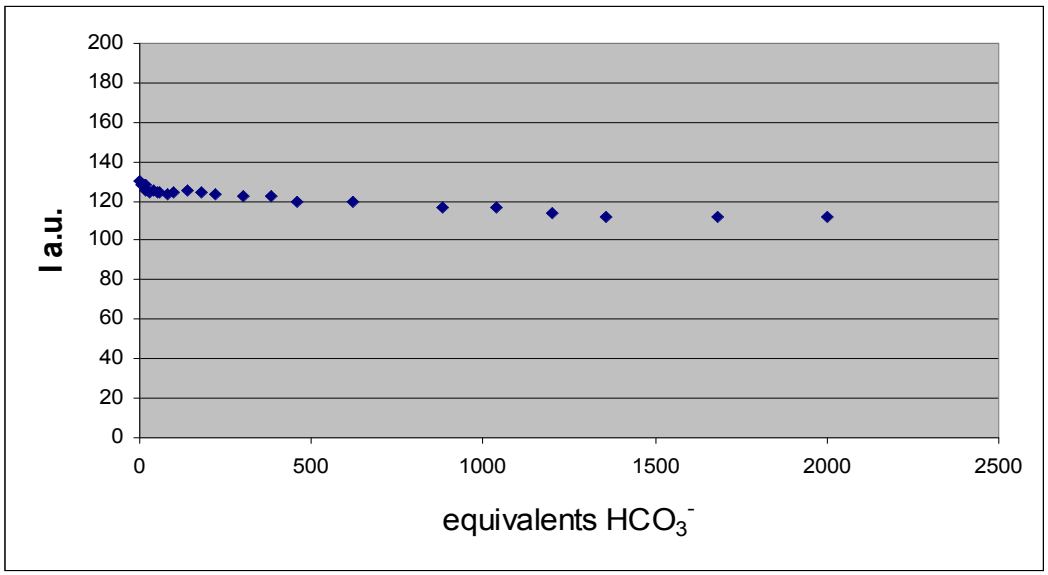
Supplementary Figure S44. Changes observed in the fluorescence spectra of **2** (10^{-5} M water DMSO 98/2 v/v, 5mM HEPES buffer pH=7.2) upon addition of NaOH in water. (λ_{ex} = 260nm)



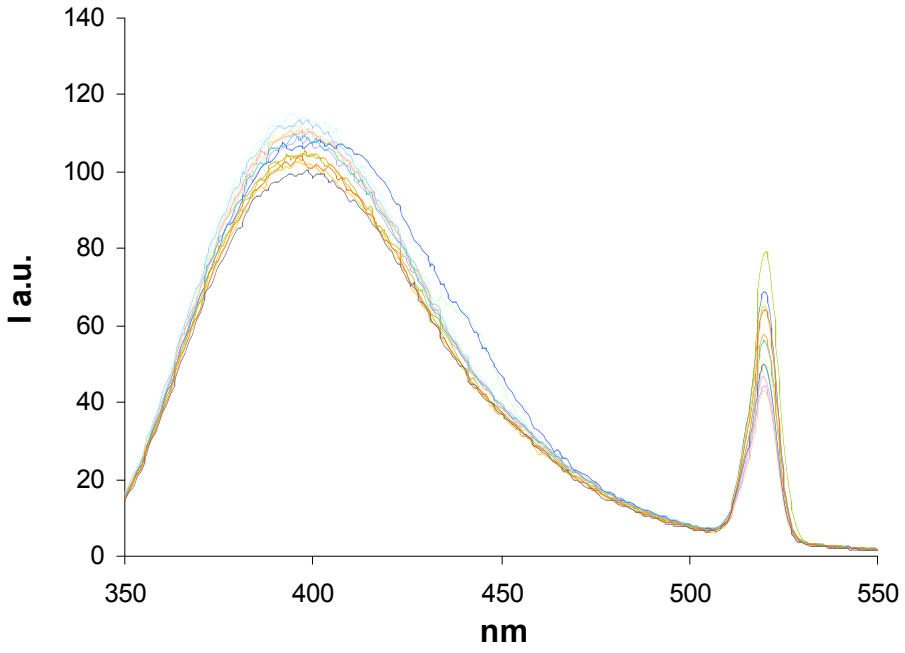
Supplementary Figure S45. Titration profile of changes observed in the fluorescence spectra of **2** (10^{-5} M water DMSO 98/2 v/v, 5mM HEPES buffer pH=7.2) upon addition of NaOH in water. (λ_{ex} = 260nm)



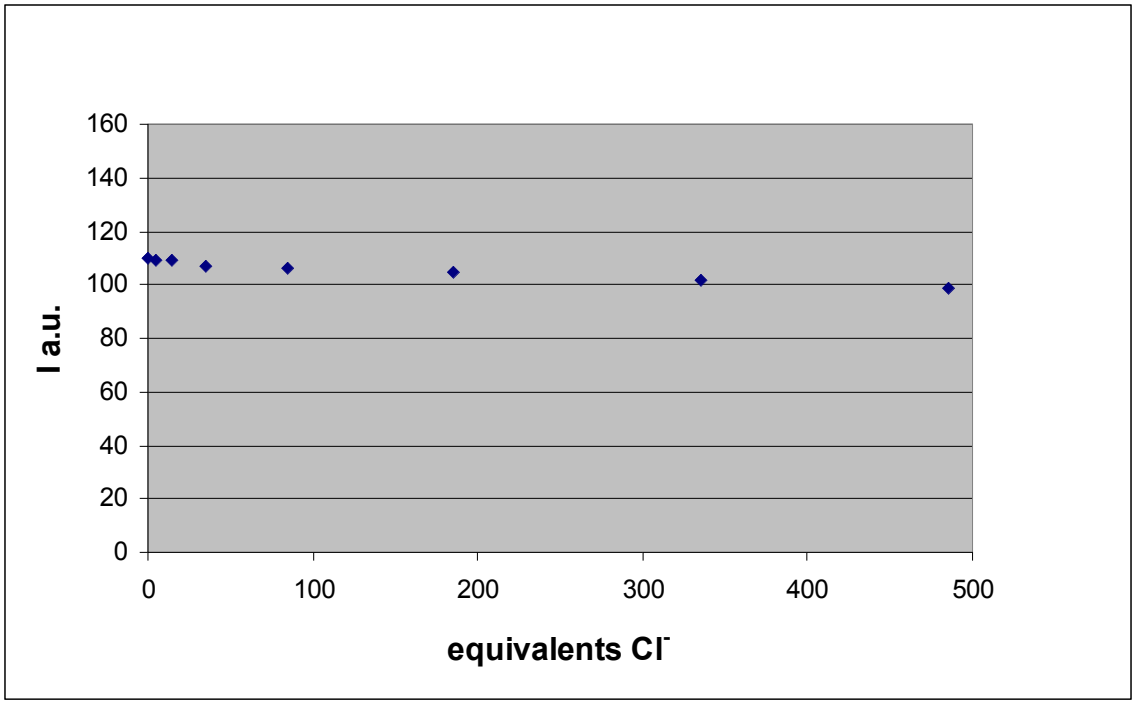
Supplementary Figure S46. Changes observed in the fluorescence spectra of **2** (10^{-5} M water DMSO 98/2 v/v, 5mM HEPES buffer pH=7.2) upon addition of $NaHCO_3$ in water. (λ_{ex} = 260nm)



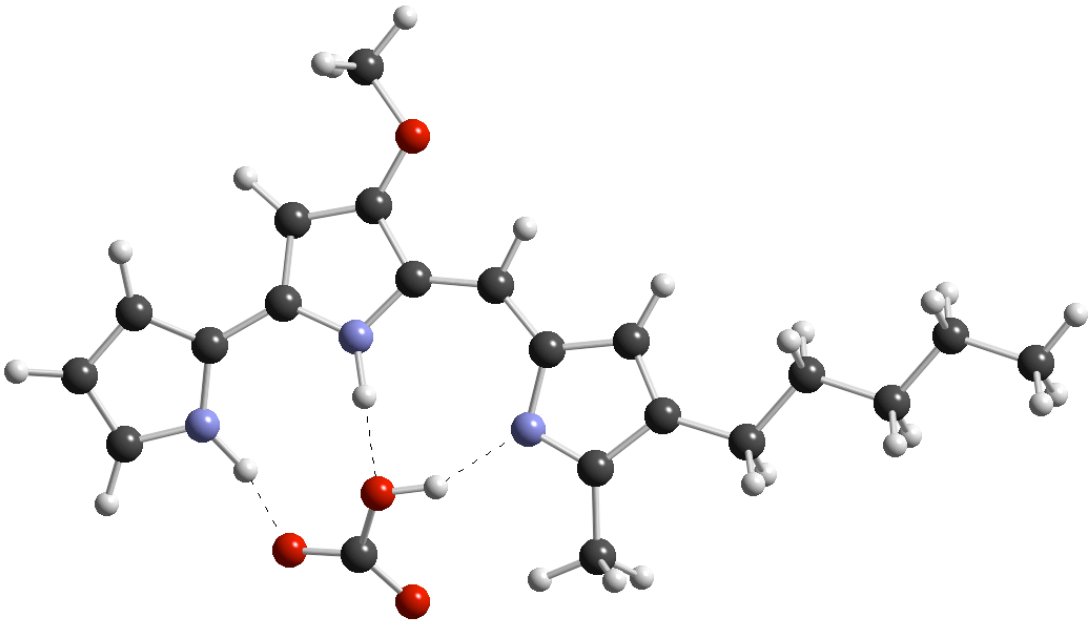
Supplementary Figure S47. Titration profile of changes observed in the fluorescence spectra of **2** (10^{-5} M water DMSO 98/2 v/v, 5mM HEPES buffer pH=7.2) upon addition of $NaHCO_3$ in water. (λ_{ex} = 260nm)



Supplementary figure S48. Changes observed in the fluorescence spectra of **2** (10^{-5} M water DMSO 98/2 v/v, 5mM phosphate buffer pH=7.2) upon addition of NaCl in water. (λ_{ex} = 260nm)



Supplementary figure S49. Titration profile of changes observed in the fluorescence spectra of **2** (10^{-5} M water DMSO 98/2 v/v, phosphate buffer pH=7.2) upon addition of NaCl in water. (λ_{ex} = 260nm)



Supplementary Figure S50 A DFT calculated structure for the complex between prodigiosin **1** and bicarbonate

Table S2: Output from the DFT calculation on MacSpartan ’06 Wavefunction Software, CA.

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Frequency Calculation
Adjusted 8 (out of 162) low frequency modes
Reason for exit: Successful completion
Mechanics CPU Time :          .09
Mechanics Wall Time:          .22
MacSPARTAN '06 Quantum Mechanics Program:  (x86/Darwin)    build  129cv3
Job type: Geometry optimization.
Method: RB3LYP Basis set: 6-31G(D)
Number of shells: 164
Number of basis functions: 472
Multiplicity: 1
SCF model:  A restricted hybrid HF-DFT SCF calculation will be  performed using Pulay DIIS + Geometric Direct
Minimization

Optimization:                 Step        Energy          Max Grad.      Max Dist.
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                              2     -1281.1042762       0.020395       0.118926
                              3     -1281.1126122       0.010160       0.130485
                              4     -1281.1166728       0.006018       0.107088
                              5     -1281.1188655       0.006910       0.093588
                              6     -1281.1202565       0.007528       0.100567
                              7     -1281.1215883       0.007275       0.089566
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                              32     -1281.1317935       0.000506       0.036178
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Quantum Calculation Wall Time:   9:13:05.3
MacSPARTAN '06 Semi-Empirical Program:  (x86/Darwin)         build  129c
Semi-empirical Property Calculation
M0001
Guess from Archive
Energy Due to Solvation
Solvation Energy SM5.4/A       -435.646
Memory Used:         12.768 Mb
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Semi-Empirical Program Wall Time:         1.14
SPARTAN PROPERTIES PACKAGE: MAC/P4            build 129c
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molecule M0001 terminated normally
End-   molecule "M0001" Mon Dec 22 22:18:36 2008
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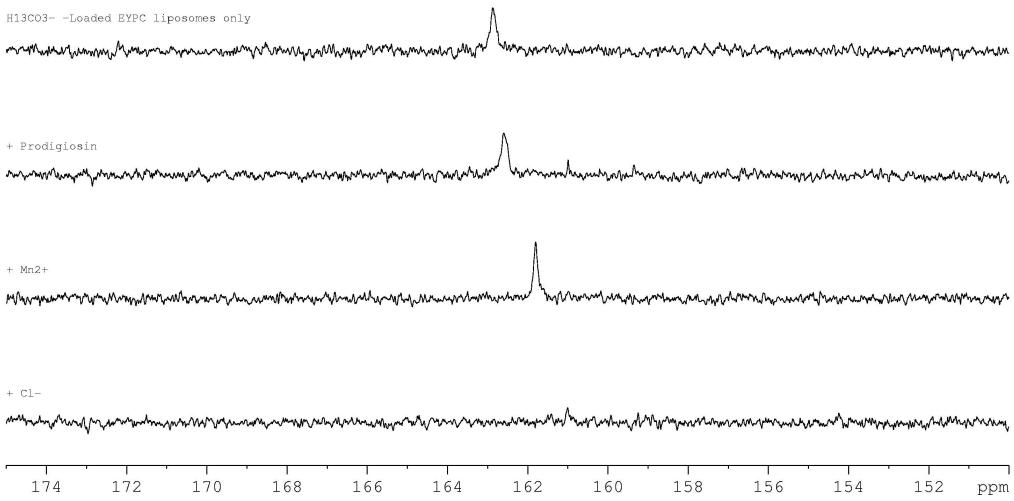
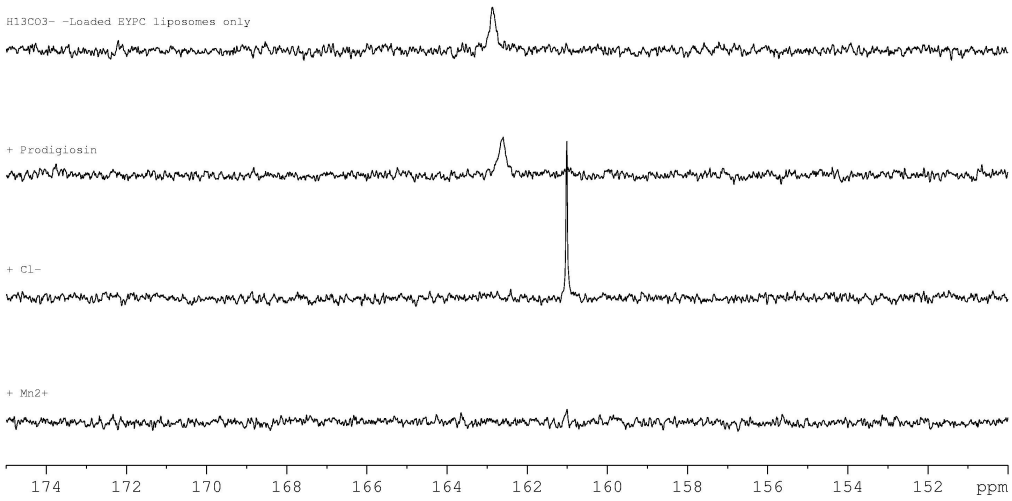
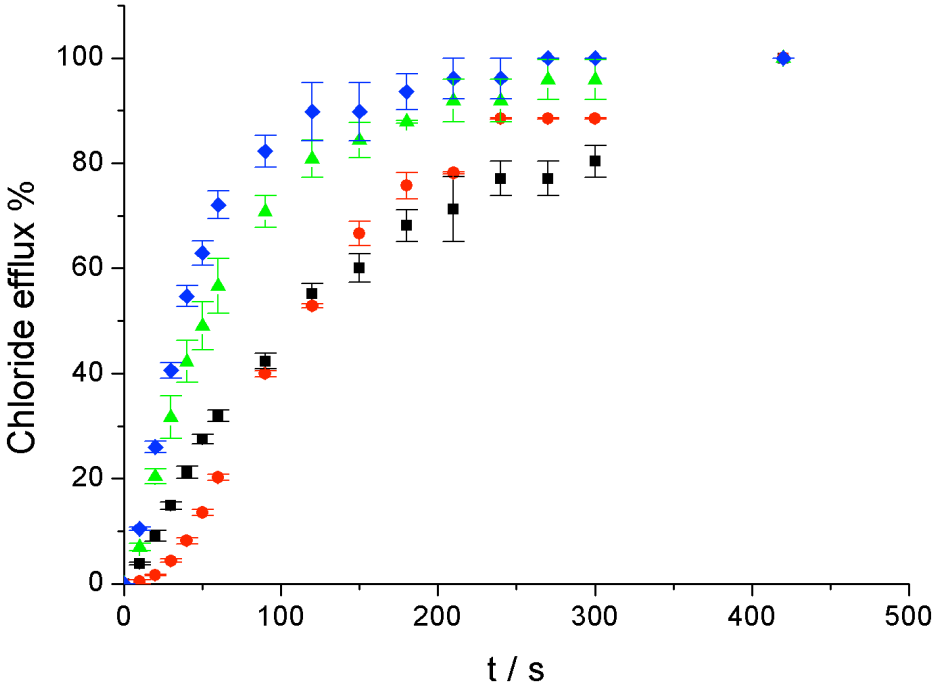


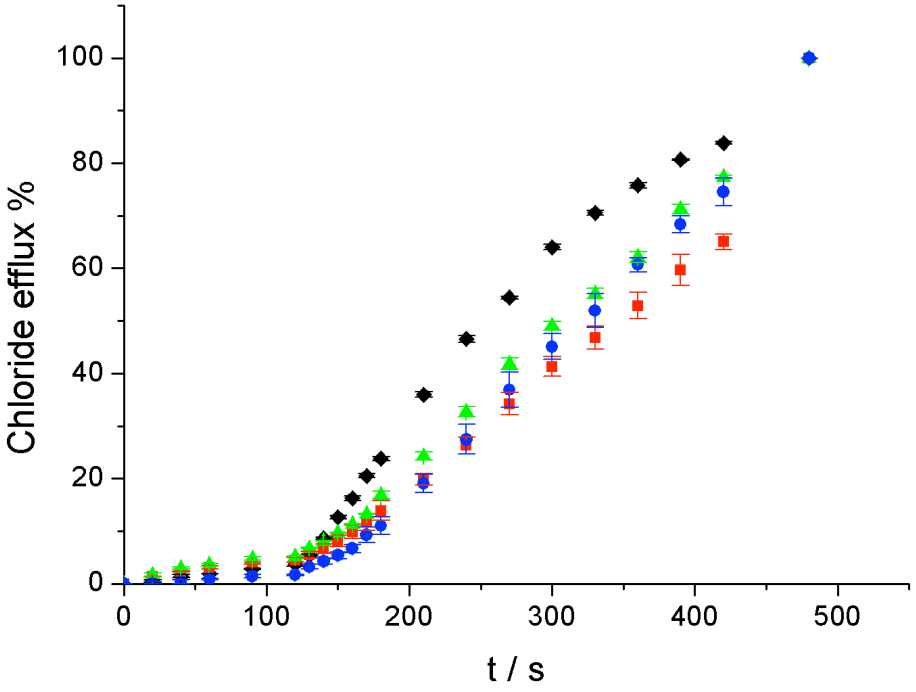
Supplementary Figure S51 Confirmation that prodigiosin does not function as a Na^+/HCO_3^- co-transporter. (From top) ^{13}C labeled HCO_3^- encapsulated inside liposomes (100 mM $NaHCO_3$, 20 mM Hepes, pH 7.3 suspended in 75 mM Na_2SO_4, 20 mM Hepes, pH 7.3). Prodigiosin added (0.1 mol%) followed by Mn^{2+} (0.5 mM). The C-13 Signal is still intact in the presence of the Mn^{2+} indicating that prodigiosin has not moved any $NaHCO_3^-$ outside of the vesicle. Upon addition of NaCl (50 mM) the C-13 NMR signal immediately disappears as prodigiosin catalyzes HCO_3^-/Cl^- exchange.



Supplementary Figure S52 (From top) ^{13}C labeled HCO_3^- encapsulated inside liposomes (100 mM $NaHCO_3$, 20 mM Hepes, pH 7.3 suspended in 75 mM Na_2SO_4, 20 mM Hepes, pH 7.3). Prodigiosin (0.1 mol%) was added: no significant shift in the ^{13}C signal was observed. Upon addition of NaCl (50 mM) the ^{13}C NMR signal shifts as prodigiosin catalyzes HCO_3^-/Cl^- exchange. Upon addition of Mn^{2+} the ^{13}C signal disappears as all the HCO_3^- has been transported out of the vesicle upon addition of chloride in a bicarbonate/chloride antiport process.



Supplementary Figure S53 A comparison of chloride efflux from synthetic vesicles mediated by compounds **1** – **4** in nitrate and sulfate solution. (a) Chloride efflux promoted upon addition of **1** (♦) (0.005 % molar carrier to lipid) and **2** (■), **3** (▲), **4** (●) (0.1 % molar carrier to lipid) to unilamellar POPC vesicles loaded with 488 mM NaCl 5 mM phosphate buffer pH 7.2 dispersed in 488 mM $NaNO_3$ 5 mM phosphate buffer pH 7.2. At t = 300 s the vesicles were lysed by addition of detergent and the final reading at t = 420 s was considered to equal 100% chloride efflux. This is the same Figure as Figure 2(a) in the manuscript but includes error bars.



Supplementary Figure S54 (a) i) Chloride efflux promoted upon addition of **1** (♦) (0.04 % molar carrier to lipid) and **2** (■), **3** (▲), **4** (●) (1 % molar carrier to lipid) to unilamellar POPC vesicles loaded with 451 mM NaCl and 20 mM phosphate buffer pH 7.2 dispersed in 150 mM Na_2SO_4 20 mM phosphate buffer pH 7.2. ii) At t = 120 s a solution of $NaHCO_3$ was added to give a 40 mM external concentration. At t = 420 s the vesicles were lysed by addition of detergent and the final reading at t = 540 s was considered to equal 100% chloride efflux. This is the same Figure as Figure 3(a) in the manuscript but includes error bars.